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Compilation of 1992 Annual Reports
of the Navy ELF Communications System
Ecological Monitoring Program

3

Volume 2 of 3 Volumes:
Tabs C-F

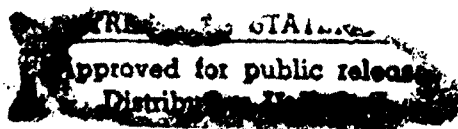
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Technical Report D06205-2
Contract No. N00039-93-C-0001
August 1993

Prepared for:

Submarine Communications Project Office
Space and Naval Warfare Systems Command
Washington, D.C. 20363-5100

Submitted by:



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13. ABSTRACT (Maximum 200 words) During 1992, the U.S. Navy continued to conduct a program to monitor flora, fauna, and their ecological relationships for possible effects from electromagnetic (EM) fields produced by the Navy's Extremely Low Frequency (ELF) Communications System. Physiological, developmental, behavioral, and ecological variables for dominant biota in upland and riverine habitats near the Naval Radio Transmitting Facility at Republic, Michigan (NRTF-Republic) have been monitored since 1982. The NRTF-Republic was intermittently energized at low amperages beginning in early 1986. Electric current and periods of energization were then gradually increased until 1989, when the transmitter became a fully operational facility. A split-plot or blocked strategy was used to examine biological variables for possible effects from EM exposure. Reports compiled in this document present the progress of these studies through 1992. It is anticipated that data will continue to be collected through 1993. Final results and conclusions are expected after all data have been analyzed in 1994. Investigators for similar studies completed in Wisconsin concluded that there were no EM bioeffects from intermittent or full operation of the transmitter in that state.				
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FOREWORD


This compendium has been prepared by IIT Research Institute (IITRI) on behalf of the Space and Naval Warfare Systems Command (SPAWAR) to document the results of studies monitoring for possible electromagnetic effects to biota from operation of the U.S. Navy's ELF Communications System.

Monitoring studies have been performed by research teams from Michigan State University, Michigan Technological University, the University of Minnesota-Duluth, the University of Wisconsin-Milwaukee, and the University of Wisconsin-Parkside under subcontract agreements with IITRI. SPAWAR funded these studies under Contracts N00039-81-C-0357, N00039-84-C-0070, N00039-88-C-0065, and N00039-93-C-0001 to IITRI. IITRI, a not-for-profit organization, managed the program and provided engineering support to ecological research teams.

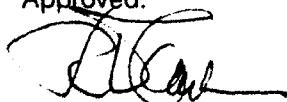
Each report in this compendium (Tabs A through H) presents the results of monitoring research performed near the Naval Radio Transmitting Facility at Republic, Michigan (NRTF-Republic) over the period 1982-1992. The results and conclusions of studies conducted near the Naval Radio Transmitting Facility at Clam Lake, Wisconsin (NRTF-Clam Lake) can be found in previous compilations. Research reports have been prepared annually, and each has been reviewed by at least three scientific peers. Investigators considered and addressed peer critiques prior to providing a final copy to IITRI for compilation. Final reports were compiled without further change or editing by SPAWAR or IITRI.

As was done for all program documents, IITRI has submitted this compilation to the National Technical Information Service for unlimited distribution. Previous compilations and other program documents are listed under Tab I.

Respectfully submitted,
IIT RESEARCH INSTITUTE


John E. Zepotosky, Ph.D.
Program Coordinator

Approved:



R. D. Carlson, Director
ELF Electromagnetic Compatibility Assurance

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IITRI D06205-1

**ELF COMMUNICATIONS SYSTEM
ECOLOGICAL MONITORING PROGRAM**

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- B. Litter Decomposition and Microflora:
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- C. Soil Amoeba:
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Novinger, D.; Kelly, B.; Rondinelli, M.; Terwin, J.
- I. Listing of Technical Reports

1. Cover page:

a. Subcontractors name and address:

Rudolph Neal Band
Department of Zoology
Michigan State University
East Lansing, MI 48824

b. Subcontract number: EO6595-88-C-003

c. Title: ELF Communications System Ecological Monitoring
Program, Task 5.2, Soil Amoeba.

d. Reporting year: November 1, 1991 to October 31, 1992.

2. Frontispage:

a. Subcontractors name and address:

Rudolph Neal Band
Department of Zoology
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East Lansing, MI 48824

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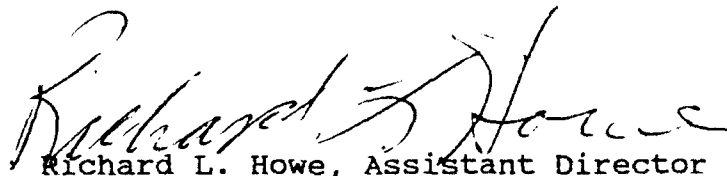
e. Name and signature of principal investigator:



Rudolph Neal Band, PI

f. Co-investigators: none

g. Name and signature of subcontractor's approving and releasing
authority:



Richard L. Howe, Assistant Director

Contract and Grant Administration

3. Table of Contents:

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4. Abstract:

Several prior years were drought years (i.e. 1986 to 1989) while 1991 was moderately dry. The as yet incomplete NOAA weather data for 1992 also indicates that this was a dry year. again growth was suppressed although not as marked as in the 1986 to 1989 drought. This is in contrast with the 1984 and 1985 growing seasons, in which abundant rainfall took place and the population increases in soil amoebae during the growing season was far greater, or the 1990 season which was intermediate in growth response.

During the 1992 growing season the ELF antenna was operational. This provided 2 years of intermittent ELF exposure (i.e. 1989 and 1990) for the biological systems to react to the radiation, one year of very limited exposure, greater exposure in 1990 and operational exposure in 1991 and 1992.

Genetic diversity data from the 1991 growing season are included in this report. The data indicated a reduction in heterogeneity at the antenna site while ground wire and control sites were similar to earlier data. This may indicate that organisms of a species vary in their response to ELF radiation depending on their genetic composition.

The antenna, ground and control sites used in previous years were continued. The sites have been characterized by IITRI personnel so that all sites have a similar 60 cycle electromagnetic background while the control site is devoid of ELF radiation from the antenna.

I have been monitoring various physical and chemical properties of the sites as well as their biological characteristics.

At all sites, population size fluctuations were observed, as was the case in previous years. Of course the fluctuations were not as dramatic for the drought seasons, as they were in 1984 and 1985.

The 1992 (and 1993) research scope was reduced from previous years so that soil counts were done in July, August and September for the organic horizon only. Instead of continuing the genetic analyses of species diversity, it was determined that an estimate of bacterial density in soil should be done. Physical and chemical properties of the soils was deleted from the 1992 study.

At all sites, changes in population size were observed, as in previous years. There was no difference in population size between antenna, ground wire and control sites.

5. Summary:

Species and strain characterization: In 1986, 1987 and 1988, Acanthamoeba polyphaga was used to test for strain heterogeneity within and between the sites. Isoenzyme analysis was used to detect strain differences; since this is a measure of genetic diversity, it is sensitive to environmental stress. No differences were found between sites. This was done again in the 1991 season, reported here, and revealed a reduction in heterogeneity at the antenna site.

Population size: the fluctuation in population of amoebae during the growing season was determined on the organic horizon for July, August and September. No differences were noted in total population size between study sites for a given horizon and sampling date. As in past years differences were noted in the number of dormant cysts between sites.

6. Progress report:

OBJECTIVES: The project objective is to determine possible effects of ELF radiation on amoebae in soil. The sites chosen for this study are adjacent to the Michigan ELF transmitter facility and include control, antenna and ground wire study sites.

In the 1991 and 1992 field season the ELF antenna was operational.

WORK PLAN ELEMENTS:

#0. Plot selection and characterization.

Synopsis: Statistical analysis of soil chemistry showed some variability between sites in past years which did not appear to affect the amoeba populations. The analysis was not done in 1992.

#1. Species and strain characterization.

Synopsis: using morphological and physiological markers, identify species and strains of soil amoebae from the study areas so that possible changes in the population due to ELF can be detected. The allozyme methods, developed for genetic analyses was used in 1986, 1987 1988 and 1991 (reported here).

#2. Population size and activity.

Synopsis: determine population size of amoebae in soil and the ratio of vegetative to dormant amoebae over the growing season. This is a productivity measure which could be affected by ELF radiation. It could also be a reflection of changes in the microbial food organisms due to ELF radiation. A new method to determine bacterial biomass has been published and was applied in the 1992 season.

Specifics: an established soil dilution counting procedure is used (Singh, 1946 as modified by Darbyshire et al., 1974). In order to count vegetative amoebae and cysts, samples are first divided in half, one-half is used to count total cysts and vegetative amoebae while the other half is treated to kill amoebae so that only cysts are counted. Differential counts are used to calculate by subtraction the total vegetative amoeba count. In the 1983 season I found that 8 random samples, subdivided into organic and mineral horizons (i.e. 8 samples per horizon), provided statistically significant data;

I will repeat this from the 1983 report: ten samples were counted from each horizon at the three sites on two dates; the results indicated a coefficient of variation that was less than 10% of the mean for a given horizon and date. From a 90% power curve, significant differences could be detected at $1.4 \times \text{std. dev.}$ for a sample size of 10 and 1.5 to $1.6 \times \text{std. dev.}$ for a sample size of 8. Thus sample sizes of 8 and 10 were almost equally powerful so that 8 random samples were taken from each horizon at the three sample sites on a sampling date.

One-way analysis of variance was used to detect differences in total amoeba and cyst count between control, antenna and ground sites for each horizon in 1989. I was advised by Professor John Gill at MSU, a statistician who works with biological problems, that log transformed data should be used for statistical analyses. Since the microbial population doubles over time, log transformed data more closely reflects biological events. Table 3 gives the error (i.e. among) degrees of freedom as 21. Direct counts of amoebae in soil, as is done with freshwater organisms (e.g. Wright & Coffin, 1984) is not possible. Microbes adhere to soil and sonication of a soil slurry to release them might make quantitative recovery of some organisms by subsequent density flotation possible, but amoebae would be destroyed.

A method to estimate bacterial biomass was developed, based on methods of Tsai & Olsen (1991) to extract bacterial DNA from soil. Quantification of extracted DNA was difficult to develop. As noted by others, including Tsai & Olsen (1991), extraction also includes DNA together with interfering substances. I tried to use a direct measurement of DNA using a fluorometer but the results were erratic due to soil contaminants. I separated the DNA from other soil substances by agarose gel electrophoresis and then quantified the DNA by ultraviolet fluorescence of ethidium bromide stained gels. Quantification was done with an "AMBIS" image analyzer. This approach is practical to do on large numbers of samples; known DNA standards run on each gel maintain accuracy. The base-line standard were laboratory cultures of

bacteria. The only reviewer to address this phase of the report felt that the work was not worth doing and separation of bacterial from fungal DNA was an open question. Tsai & Olsen (1991) did demonstrate that fungal DNA was not present in a large enough quantity to be a problem. I dropped this line of research.

#3. Data analysis.

Synopsis: statistical analyses mentioned earlier are summarized here. For amoeba counts in soil, by soil dilution procedures, a one-way analysis of variance with 8 replicates per cell was adequate (Table 3). For isoenzyme determinations, comparisons between isolates are done by the method of Nei (1972).

EXPERIMENTAL

Methods and results will be presented in reference to the Work Plan, given above.

#1. Species and strain characterization. The isoenzyme analysis of genetic heterogeneity of A. polyphaga, was done in 1991. Further analysis of this data was needed so that it is presented in this year's annual report.

The experimental design for 1991 was to isolate 10 clones of Acanthamoeba polyphaga from each of the study sites (i.e. antenna, ground wire and control) to analyze allozyme patterns from each. This was done in 1986, 1987 and 1988. The organism was chosen because cyst morphology makes this amoeba easy to identify in the plate cultures used to enumerate amoeba numbers, and it is present in reasonable numbers although not overly abundant.

During analysis of the allozyme data I discovered that the technician did 10 or more clones from the control and ground wire sites but only 7 from the antenna site. I had her isolate 10 additional clones from the antenna site to use for this analysis. I am still in the process of analyzing variation in individual enzyme loci for each site to identify the affected loci; this data will be included in the final version of this report. Table 5 gives the statistics of the genetic heterogeneity at each site and comparisons between sites. Note that data for Control and Ground Wire sites did not differ between each other or for 1986,

1987 and 1988 data for all three sites. Table 5a gives the data for Nei's genetic distance and Table 5b gives the genotypes at the Control site. Allozymes used are listed in Table 5c.

Although natural populations of the organism may or may not have a sexual reproductive phase, clones isolated from soil exhibit no sexual reproduction (i.e. selfing) and remain stable in terms of allozyme patterns over many generations. In 1989, 1990 and 1991 clone isolates were grown in culture containers subjected to ELF voltages in situ (referred to as "T-tube" cultures in the annual reports for these years). At the start of each season allozyme determinations were made on the clone for identification purposes, and the clones were grown in 18 T-tubes at the research sites. After three months of continuous subcultures, all 18 cultures were again analyzed for allozyme patterns. No changes were observed in allozyme patterns for a given clone which indicated that sexual reproduction did not take place (i.e. selfing) in a given clone. This data was presented in the annual reports for 1989, 1990 and 1991. The reviewer who addressed this aspect of the research recommended that the 1993 allozyme study be done on 30 clone isolates per site; this will be done.

#2. Population size and activity. As stated in previous annual reports, the number of replicate soil samples required to statistically compare soil amoeba populations between study sites was 8. In 1984 and 1985 soil amoeba populations increased from the start of the growing season to a peak in excess of a million amoebae/gram soil in August and then dropped sharply in September

to a few thousand/gram soil. Vegetative amoebae formed a significant component of each monthly sample, including the smaller September and October populations. No differences were noted for a given soil horizon between the antenna, ground and control sites. The drought, beginning in 1986, has had a pronounced effect on population size, the ratio of vegetative to dormant cysts and some site differences in 1987 (the June and July counts. The results from the 1990 season show population sizes characteristic of a dry year but better than the prior 4 years. In 1991 and 1992 the population size data resembled the 1986 to 1989 dry seasons. Table 1 gives total counts of vegetative amoebae and cysts while Table 2 gives counts of cysts alone, thus the mathematical difference gives the number of vegetative amoebae present in a sample. Figure 1 interprets Tables 1 and 2 in showing total counts and the calculated percent vegetative amoebae by horizon and site at various sampling dates. Figure 3 summarizes maximum average yields for all sites by year and month to illustrate the general trend in maximum population changes. Figure 4 replots this data as the average maximum yield per year to illustrate the annual trends that correlate to rainfall data. Table 3 demonstrates no significant differences between sites for a given horizon and sampling date. As in past years cyst counts exhibited differences between sites which reflect the susceptibility of vegetative and cyst states to local conditions (e.g. moisture, food) (Table 2 & 3).

I have summarized the NOAA Climatological Data publications

for monthly deviations from normal rainfall for 1985 to date (Fig. 5 & 6) to illustrate the drought years. Soil moisture measurements indicate slightly drier soils during this period (Fig. 4), which may account for the effects of the drought on growth, although nutrient input from surface litter may be a more important component of limiting amoeba growth and would correlate with the rainfall pattern. Note that both monthly rainfall (Fig 5) and soil moisture (Table 4) indicated that August was a dry month. Although in past years August frequently coincided with maximum population sizes, this was not the case in 1991 or 1992.

7. Peer reviewers and publications:

I plan to use the following individuals as peer reviewers:

a. Prof. Thomas J. Byers

Department of Molecular Genetics

Ohio State University

b. Prof. Frederick L. Schuster

Department of Biology

Brooklyn College

Publications (1991):

1. Hu, Wang-nan, Kopachik, W., Band, R.N. 1992. Cloning and characterization of transcripts showing virulence-related gene expression in Naegleria fowleri. Infect & Immun. 60, 2418-2424.

2. _____. 1992. A simple, rapid method to create a cDNA library. Biotechniques 13, 862-864.

3. Submitted for publication: Codon usage and sequence analysis in Naegleria fowleri actin cDNA. Gene.

4. Manuscript in preparation on the effect of extremely low frequency electromagnetic emissions on the genetic heterogeneity of a natural population of soil amoebae.

5. Attended the VI International Conference on the Biology and Pathogenicity of Free-Living Amoebae at the Medical College of Virginia, Richmond, August 2-7, 1992. I chaired the session on Molecular Biology and presented a paper from platform on Virulence-Related Gene Expression in Naegleria fowleri.

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1974. A rapid micromethod for estimating bacterial and protozoan
populations in soil. *Rev. Ecol. Sol* 11, 465-475.

Nei, M. 1972. Genetic distance between populations. *Am. Nat.*
106, 283-292.

Singh, B.N. 1946. A method of estimating the numbers of soil
protozoa, especially amoebae, based on their differential feeding
on bacteria. *Ann. Appl. Biol.* 33, 112-119.

Tsai, Y-L & Olson, B.H. 1991. Rapid method for direct
extraction of DNA from soil and sediments. *Appl. & Env. Micro.*
57, 1070-1074.

TABLE 1. Total counts from 8 samples per site (organic horizon):

SITE	HORIZON	DATE	MEAN #/g soil \pm S.D.* (log#)	MEAN**	C.V.***
Control	Organic	7/20	4.7251 \pm 0.2418	61,851	5.1
		8/26	3.8665 \pm 0.0804	7,471	2.1
		9/23	4.0538 \pm 0.1721	12,095	4.3
Antenna	Organic	7/20	4.5317 \pm 0.4853	58,538	10.7
		8/26	4.0124 \pm 0.1760	11,233	4.4
		9/23	4.0847 \pm 0.2482	13,799	6.1
Ground	Organic	7/20	5.0294 \pm 0.4079	168,600	8.1
		8/26	3.9838 \pm 0.1151	9,550	2.9
		9/23	3.9872 \pm 0.1341	9,839	3.4

* Mean expressed as \log_{10} number, used to calculate analysis of variance (Table 4B).

** Mean calculated from arithmetic data which will differ from converting the mean of log data to an arithmetic figure. The log of the arithmetic mean will not be the same as the mean of the log transformed data.

*** Coefficient of Variability, %.

TABLE 2. Cyst counts from 8 samples per site (organic horizon):

SITE	HORIZON	DATE	MEAN #/g soil \pm S.D.* (log#)	MEAN**	C.V.***
Control	Organic	7/20	4.4462 \pm 0.4501	44,103	10.1
		8/26	3.5328 \pm 0.1389	3,570	3.9
		9/23	3.7050 \pm 0.1241	5,250	3.4
Antenna	Organic	7/20	4.1944 \pm 0.2447	17,992	5.8
		8/26	3.5038 \pm 0.0984	3,261	2.8
		9/23	3.6648 \pm 0.0982	4,732	2.7
Ground	Organic	7/20	4.6858 \pm 0.2941	60,020	6.3
		8/26	3.5603 \pm 0.1445	3,428	4.1
		9/23	3.6934 \pm 0.1129	4,724	3.1

* Mean expressed as \log_{10} number, used to calculate analysis of variance (Table 4B).

** Mean calculated from arithmetic data which will differ from converting the mean of log data to an arithmetic figure. The log of the arithmetic mean will not be the same as the mean of the log transformed data.

*** Coefficient of Variability, %.

TABLE 3. One-way analysis of variance by date and horizon. Data log transformed (see Table 1 & 2).

HORIZON	DATE	GROUPS	DF	TOTAL COUNT		F	POWER
					MS		
ORGANIC	7/20	among	2	0.5036	3.2812	NS	0.57
		within	21	0.1535			
	8/26	among	2	0.0474	2.7747	NS	0.5
		within	21	0.0171			
	9/23	among	2	0.0183	0.4909	NS	0.13
		within	21	0.0373			
CYST COUNT							
ORGANIC	7/20	among	2	0.4831	4.1522	*	(0.995)
		within	21	0.1163			
	8/26	among	2	0.0068	0.4026	NS	0.1
		within	21	0.0168			
	9/23	among	2	0.0034	0.2720	NS	0.09
		within	21	0.0126			

* = 5% significance level

** = 1% significance level

TABLE 4. SOIL MOISTURE (% W/W)¹, ORGANIC HORIZON.

	CONTROL SITE	ANTENNA SITE	GROUND SITE
DATE			
JUL 20	48±13	48±9.5	49.8±7
AUG 26	27.4±6.9	33.3±2.7	28.3±7
SEP 23	37.4±7	36.1±4	37.3±9.7

ONE WAY ANOVA (between sites)

Date	D.F.	M.S.	F
JUL20 Between	2	6.8	0.065
Within	21	103.9	
AUG26 Between	2	80.37	2.54
Within	21	35.7	
SEP23 Between	2	3.8	0.072
Within	21	52.9	

¹ = mean ± S.D. (n=8)

TABLE 5. Genetic diversity expressed as genetic distance.

1. Summary data:

SITE	Mean genetic distance \pm Std. Dev.			
	1986	1987	1988	1991
Control	0.5108 \pm 0.175	0.5494 \pm 0.175	0.5643 \pm 0.210	0.5713 \pm 0.228
Antenna	0.5589 \pm 0.178	0.5558 \pm 0.198	0.5333 \pm 0.147	0.3154 \pm 0.119
Antenna #2, for 1991				0.4459 \pm 0.1562
Ground	0.5314 \pm 0.186	0.5229 \pm 0.167	0.5026 0.129	0.5878 \pm 0.168

2. 1991 statistics:

One-way ANOVA:

	d.f.	M.S.	F
Among	2	1.04938	33.23546
Within	132	0.03157	

P value < 0.0001, significant difference

Bonferroni t-test:

Comparison	Mean Difference	uncorr. p value	Bonferroni p value
Control vs. Antenna	0.2559	<0.0001	***p<0.001
" vs. Ant#2	0.1254	0.0009	**p<0.01
Control vs. ground	-0.0165	0.6508	NS
Antenna vs. ground	0.2724	<0.0001	*** p<0.001
Ant#2 vs. "	0.1419	0.0002	**p<0.01

3. Multiyear comparisons (1X ANOVA per site):

CONTROL	d.f.	M.S.	F
Among	3	0.03286	0.835434 NS
Within	176	0.03934	
ANTENNA			
Among	4	0.48432	18.47746 **
Within	220	0.02621	
GROUND			
Among	3	0.05987	2.230847 NS
Within	176	0.02684	

Unpaired t-test for 1988 antenna and 1991 antenna #2: two-tailed p value is 0.0076 (very significant difference).

Table 5a.

Isozyme analysis, 1991 season. Nei's Genetic Distance.

FROM ANTENNA SITE, 60 alleles at 27 loci										
	1	2	3	4	5	6	7	8*	9*	10*
1	-	.218	.162	.359	.235	.516	.345	.503	.414	.51
2		-	.223	.336	.523	.381	.227	.728	.427	.544
3			-	.297	.317	.632	.264	.563	.499	.544
4				-	.359	.524	.322	.659	.661	.599
5					-	.298	.282	.667	.464	.538
6						-	.436	.78	.601	.51
7							-	.611	.51	.614
8								-	.362	.5
9									-	.287
10										-

* Clones 8, 9 & 10 added to original 7.

mean = 0.4459; s.d. = 0.1562

FROM ANTENNA SITE, 51 alleles at 24 loci										
	1	2	3	4	5	6	7	8	9	10
1	-	.206	.155	.214	.273	.364	.499	.456	.379	.38
2		-	.292	.396	.34	.182	.298	.261	.265	.179
3			-	.279	.244	.358	.413	.547	.398	.374
4				-	.331	.449	.537	.413	.338	.465
5					-	.242	.425	.464	.482	.409
6						-	.138	.122	.206	.179
7							-	.154	.277	.293
8								-	.184	.256
9									-	.079
10										-

mean = 0.3154; s.d. = 0.1192

Table 5a cont'd.

Isozyme analysis, 1991 season. Nei's Genetic Distance.

FROM CONTROL SITE, 60 alleles at 27 loci

	1	2	3	4	5	6	7	8	9	10
1	-	.268	.63	.559	.428	.467	.513	.515	.733	.412
2		-	.456	.372	.523	.516	.523	.554	.714	.537
3			-	.471	.685	.514	.593	.513	1.28	.637
4				-	.559	.506	.405	.39	1.02	.365
5					-	.698	.438	.521	1	.614
6						-	.429	.403	.832	.384
7							-	.207	1	.35
8								-	1.04	.21
9									-	.987
10										-

mean = 0.5713; s.d. = 0.2282

FROM GROUND SITE, 66 alleles at 27 loci

	1	2	3	4	5	6	7	8	9	10
1	-	.5	.169	.515	.413	.577	.331	.726	.869	.573
2		-	.602	.688	.685	.588	.522	.527	.397	.557
3			-	.517	.411	.554	.381	.586	.724	.435
4				-	.68	1.05	.822	.887	.887	.659
5					-	.334	.566	.547	.844	.549
6						-	.652	.524	.787	.574
7							-	.564	.538	.648
8								-	.49	.456
9									-	.543
10										-

mean = 0.5878; s.d. = 0.1687

Table 5b.

Genotypes at control site, 1991:

locus	1	2	3	4	5	6	7	8	9	10
LDH 1	1	1	1	1	1	1	1	1	0	0
LDH 2	1/2	1/2	1/3	1	2	1/3	1/2	1/2	0	1/2
LTD 1	0	0	1	0	0	1	0	1	0	1
LTD 2	1/2	1/2	1	0	0	1	0	1	0	1
HK 1	2	2	2	0	0	0	0	0	0	0
PE 1	0	0	1	0	2	0	0	1	0	0
PE 2	0	0	1/2	0	0	0	1	1/2	0	2
PE 3	1/2	0	1	0	0	1/3	1/3	1	0	2
PE 4	3	2/3	2	2/3	2/3	1/3	2/3	2/3	0	1/3
PE 5	1/2	1	1	1	1/2	1	1/2	1	0	1
BE2 1	0	1/2	1/2	1	1	0	0	0	0	0
BE2 2	3	3	1/3	3	3	1/3	1/3	1/3	0	1/3
BE2 3	1/2	1/2	1	2	0	1	1/2	1/2	0	1/2
BE2 4	1/3	1/2	1/2	2	1	0	1	0	0	0
BE2 5	1	0	0	0	1	1/2	2	1/2	0	1/2
BE2 6	2	2	0	1/2	1/2	1/2	1/2	1/2	2	1/2
ICD 1	0	0	0	1/2	0	0	1/2	0	0	0
ICD 2	2	0	2	2	0	0	0	0	2	0
ICD 3	0	1/2	2/3	1	0	1/2	1	1	0	1
ICD 4	1/2	1/2	1/3	2/3	2/3	0	1/2	2	0	2
AE 1	2	3	1/2	1/2	1/2	1/2	1/2	1/2	0	2
AE 2	1/3	2/3	1/3	0	2/3	3	1	0	2/3	2/3
AE 3	1/3	1/3	2/3	2/3	2/3	3	3	3	1/3	1/3
AE 4	1/2	1/2	1/2	1/2	1	1	1/2	1/2	0	1/2
PGM 1	0	0	1	1/2	0	0	0	2	0	1/2
PGM 2	0	0	0	1	1	0	0	1	0	1
PGM 3	0	0	2	2	2	0	1/2	1/2	0	1/2

Table 5c.

Allozymes assayed for 1991:

Lactate dehydrogenase (LDH, 2 loci)

Threonine dehydrogenase (LTD, 2 loci)

Hexokinase (HK, 1 locus)

Propionyl esterase (PE, 4 loci)

Butyryl esterase-2 (BE, 6 loci)

Isocitrate dehydrogenase (ICD, 4 loci)

Acetyl esterase (AE, 4 loci)

Phosphoglucomutase (PGM, 3 loci)

Figure 1. Summary of soil counts, 1992.

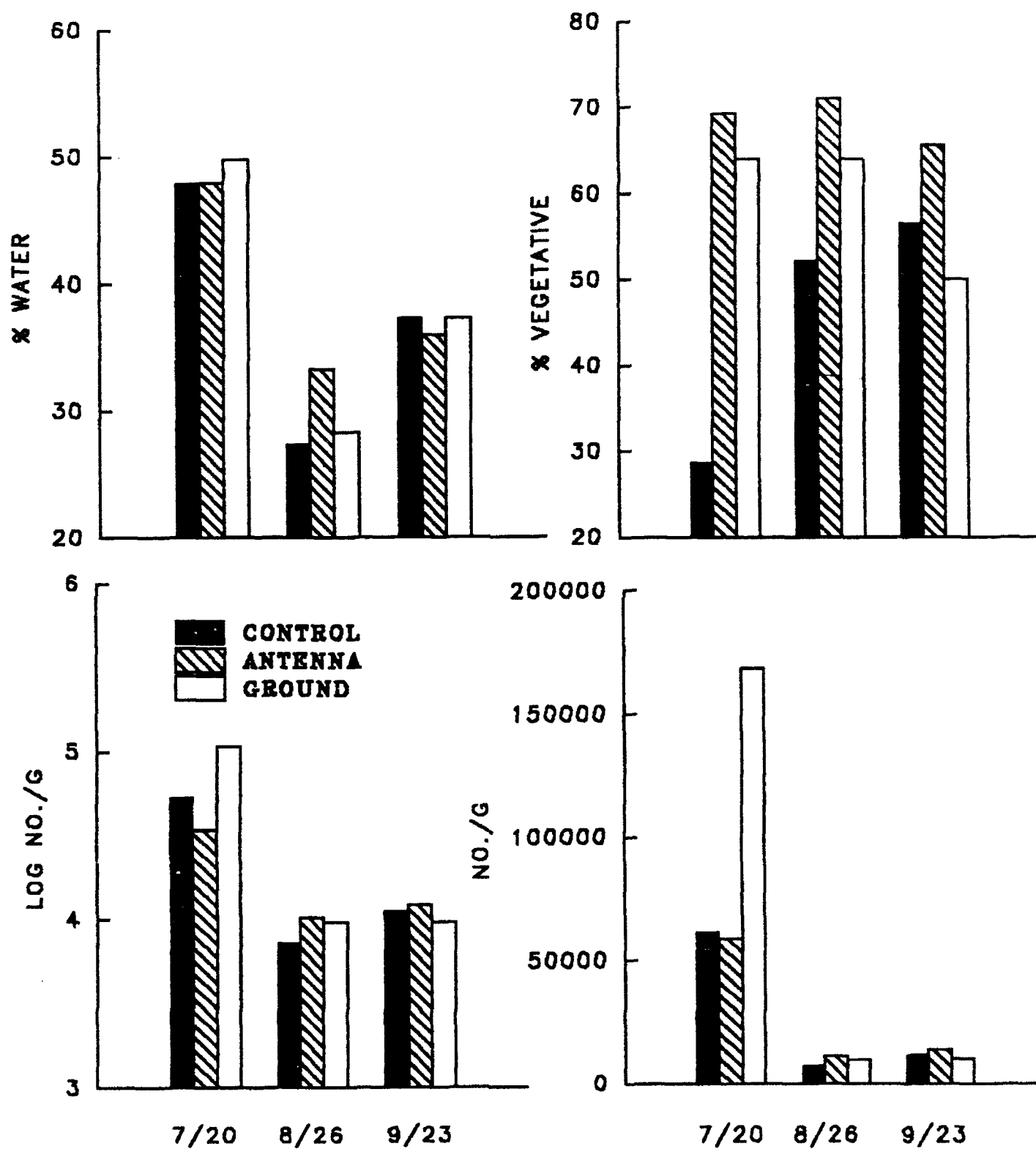


Figure 2. Average yields by month and year for all sites.

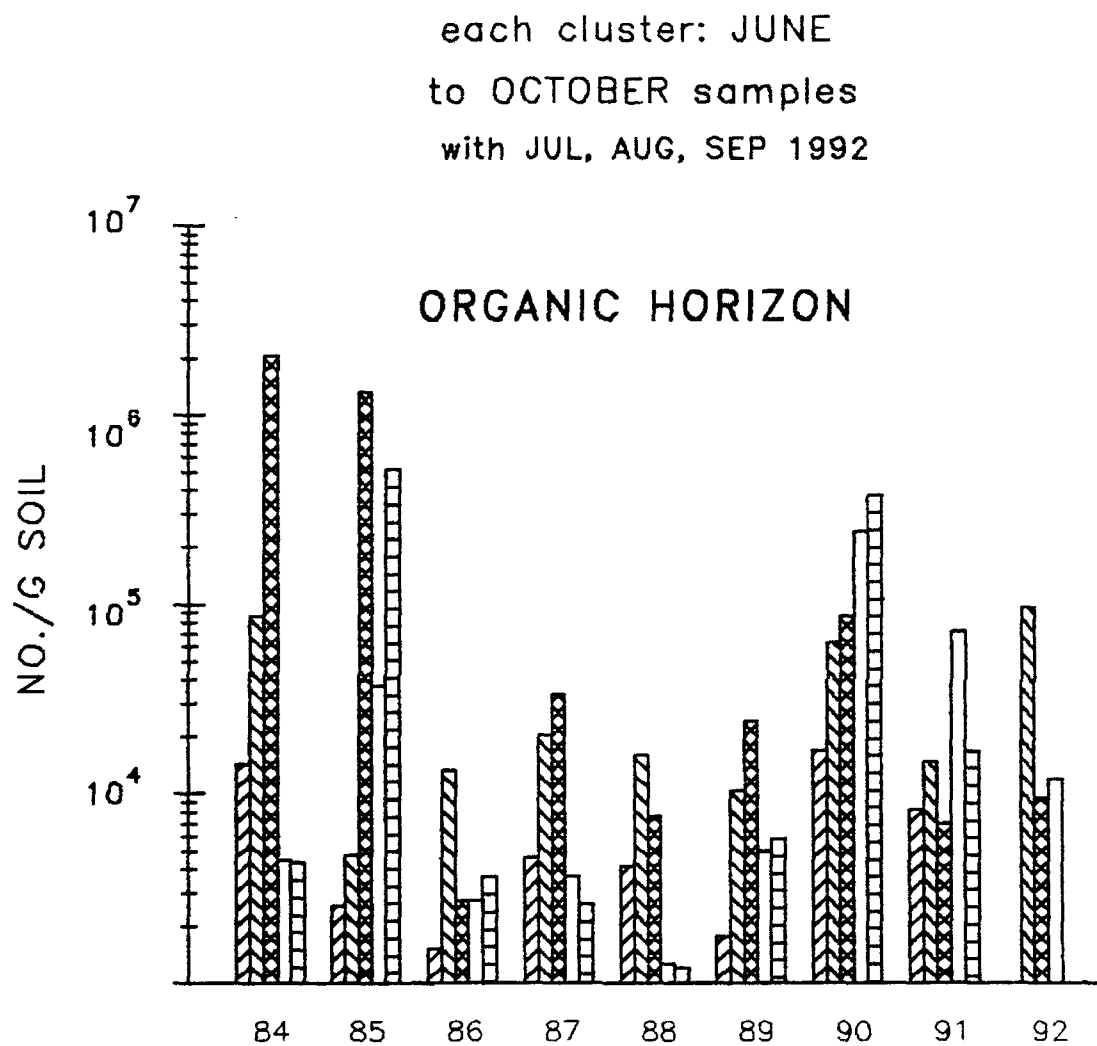


Figure 3. Average maximum total amoebae per year.

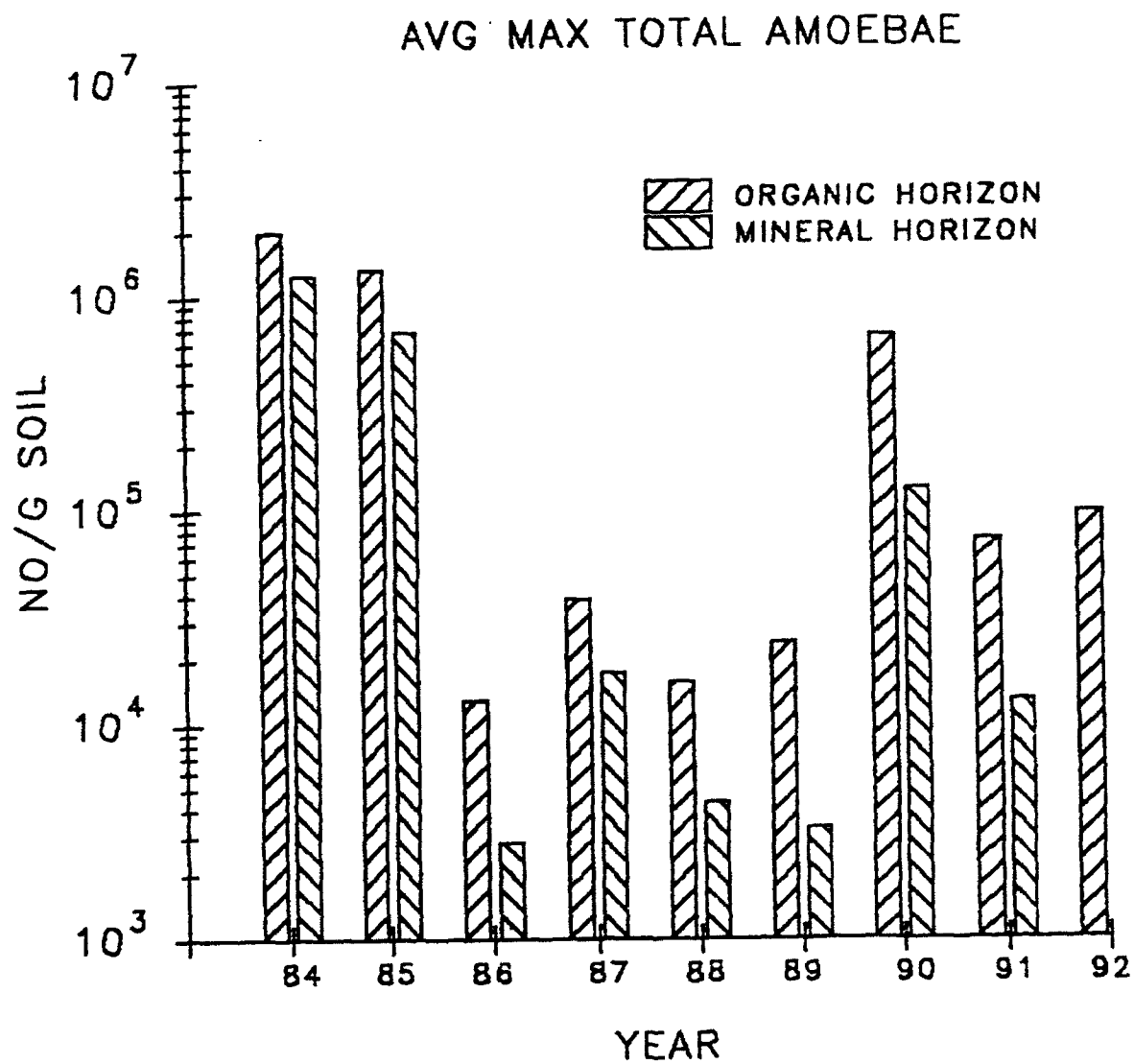


Figure 4. Mean soil moisture of samples taken for counting amoebae.

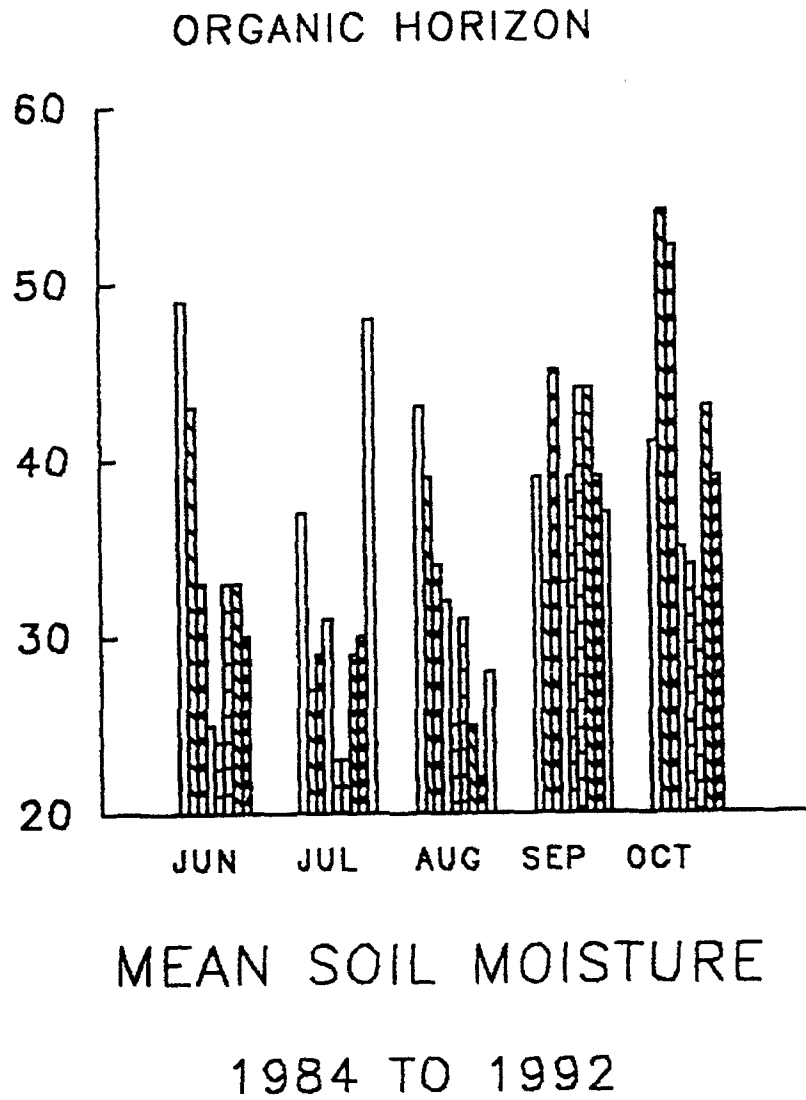


Figure 5. Annual rainfall departure from normal for all years.

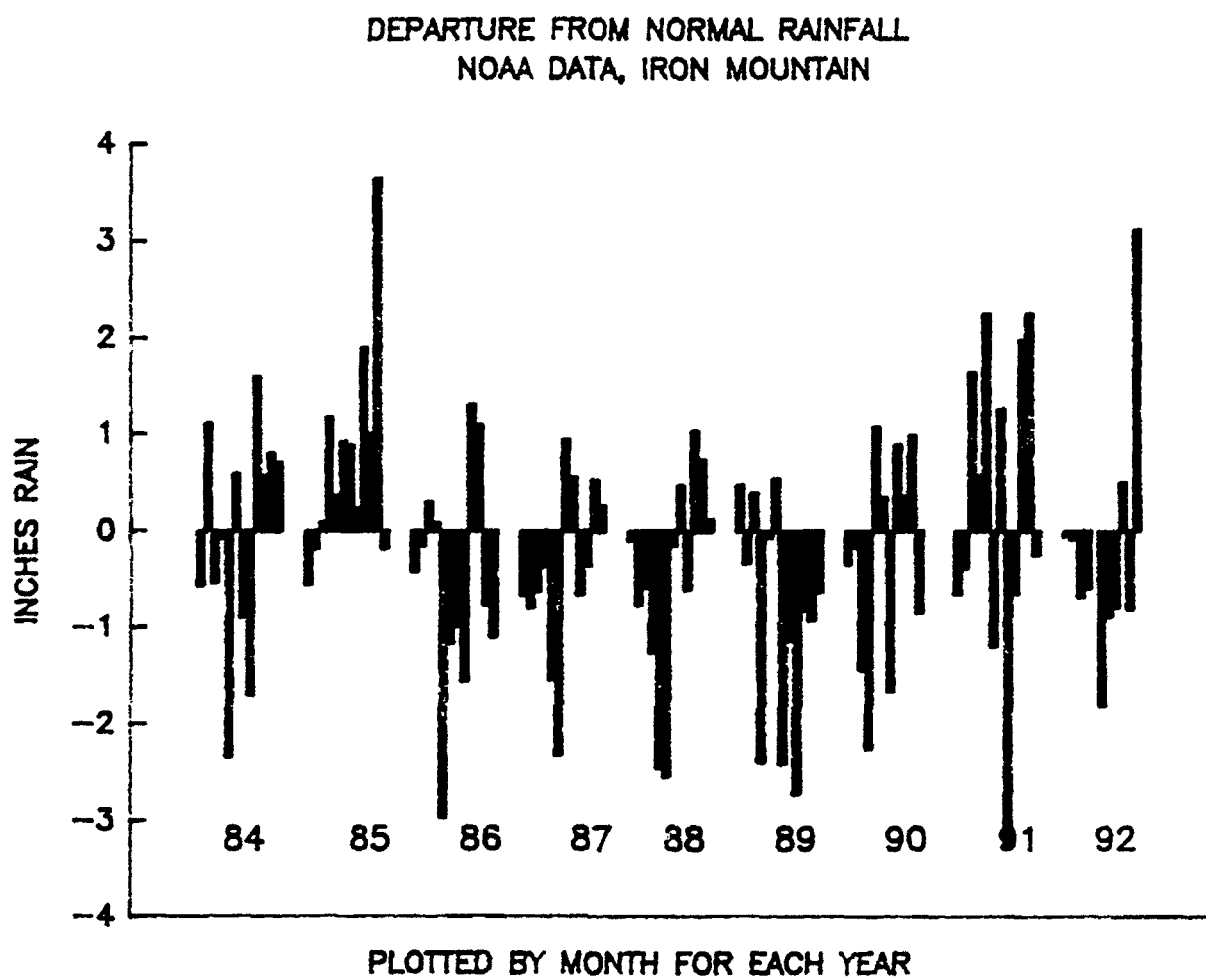
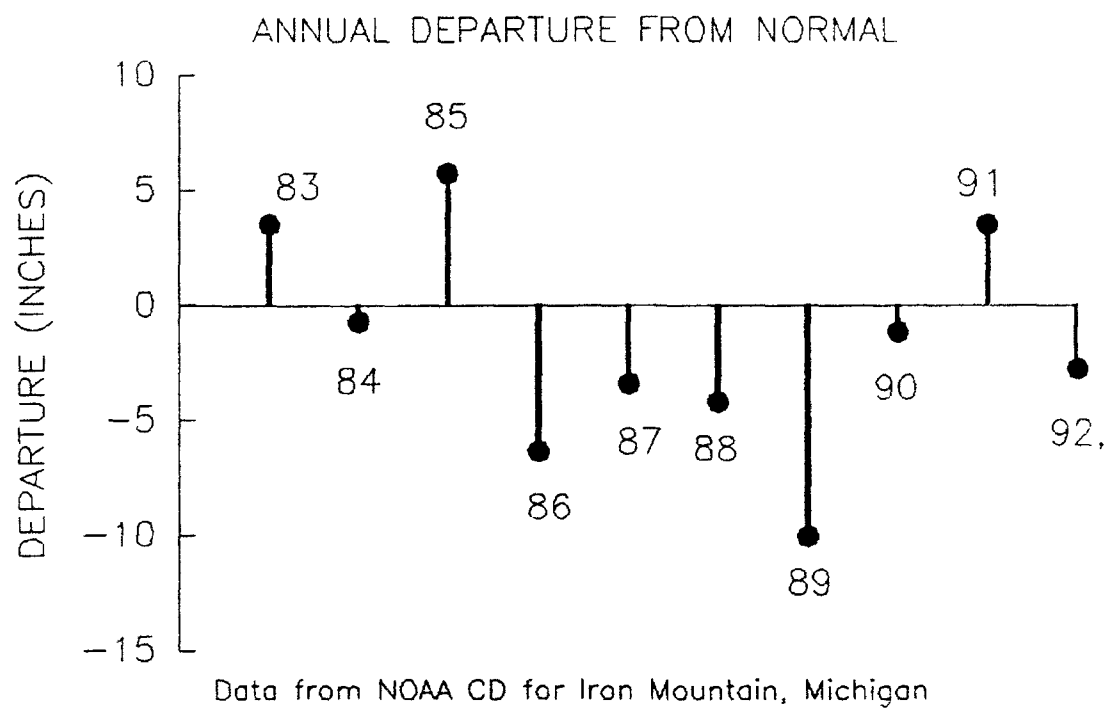


Figure 6. Annual departure from normal rainfall.



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ELF Communications System Ecological Monitoring Program
Arthropoda and Earthworms
Tasks 5.3. and 5.4.

ANNUAL REPORT

1992

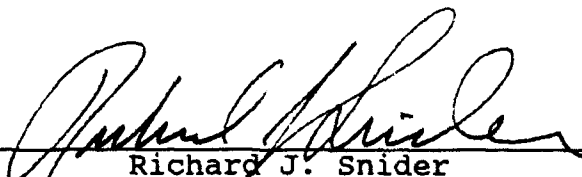
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Arthropoda and Earthworms

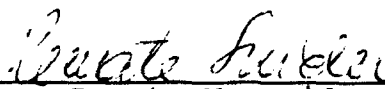
Tasks 5.3. and 5.4.

ANNUAL REPORT

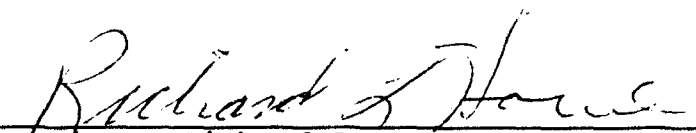
1992



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ABSTRACT

We define the pre-operational period as spanning 1984-88, except for elements in which the methodology was not finalized until 1985 (surface-active arthropods) or 1986 (most soil-dwelling arthropods). Depending on the status of progress in each work element, data are now available for three or four operational years, beginning with 1989.

Most soil- and litter-dwelling arthropods showed no detectable response to antenna operation. Where statistically significant differences between pre-ELF and operational data existed, they could not be interpreted as effects of EM fields. Rather, long-term numerical fluctuations (often associated with the Control and not the Test site) seemed to be the underlying cause.

Seasonal activity patterns of Collembola, velvet mites and carabid beetles showed no significant deviations in 1989 through 1991, the last year in which pit-trapping was performed. Patterns of arthropod development, measured by the seasonal appearance of life stages, were equally unaffected.

For the dominant soil-dwelling earthworm species in the Test site, we detected subtle effects of EM fields. The population exhibited reduced reproductive activity; seasonal fluctuations (which are normally strongly influenced by soil moisture) were no longer clearly dependent on environmental conditions. Through use of retrievable mesh bags in which groups of earthworms were isolated, it was determined that rates of cocoon production were increased during exposure to EM fields.

Inputs of leaf litter from the forest canopy and seasonal fluctuations of litter mass on the forest floor remained consistent with pre-operational observations. Although 1992 data on litter breakdown rates are not yet analysed, available information indicates that no EM field effects are detectable with respect to any of the system-level parameters monitored.

SUMMARY

Sampling intervals and methods of previous years were adhered to in 1992, from May 12 through October 12. Earthworms, litter and soil moisture, and litter and soil arthropods were sampled at intervals of 2 weeks; temperature, rainfall, litter input and decomposition data were obtained at intervals appropriate to each study element. Earthworm isolation experiments, using fiberglass mesh bags embedded in the soil, were continued with expanded replication. For these experiments, a technique for non-destructive assessment of soil moisture (Time Domain Reflectometry) was validated.

All arthropod data bases are available through 1991. In the case of surface-active arthropods, 1991 was the final year of study; for litter and soil arthropods, 1992 samples are still being processed. Information on earthworm phenology, earthworm isolation experiments, and litter inputs is available through 1992.

Data bases were divided into a pre-ELF (all years through 1988) and an operational (1989 onward) period. Statistical analyses were performed as appropriate, depending on the nature of the data base, the distribution of the data and their variances. Most commonly, BACI (Before and After Control and Impact) or Kruskal-Wallis procedures were employed. For variables unique to the Test site (some earthworm parameters), regression models were developed using various independent variables. For earthworm isolation experiments, ANOVA was used whenever possible.

In keeping with previous reports, the following summary is divided into three main categories:

1. Seasonal and yearly fluctuations in population abundance:

Most arthropod species exhibited no clear numerical changes following ELF activation. Where statistically significant differences existed, they were usually attributable to changes in Control rather than Test populations. We believe that what we are recording are very long-term fluctuations in species abundances, which would continue to produce occasional significant differences if further monitoring were to be pursued over several more years. There is no evidence that these changes are related to antenna activation.

Data on earthworm population abundance provide an overview parameter useful for understanding long-term trends. Densities, however, are the end result of specific phenological events, which in turn are greatly affected by climatic patterns. It is these events which must be the subject of detailed analysis.

2. Population parameters other than abundance per se:

a. Community composition and structure: Diversity and equitability indices are commonly used descriptive community parameters. For both soil/litter and surface-active arthropods, occasional significant differences between pre-ELF and operational years were detected. However, a closer look at the data from which these indices are derived led to the conclusion that these differences were of no biological significance, and were not ELF-related. Total numbers of individual species (the basis for

community indices) can vary drastically from year to year; for soil Collembola, for instance, an unusual decrease in dominant species was observed in the Control site, but not in Test; in several instances, reversal of pre-ELF relationships between Test and Control data began in 1988, prior to ELF activation, and thus independent of it.

b. Population structure: Of three species of soil or litter arthropods in which we distinguish developmental stages, none exhibited detectable effects of antenna operation. Pit-trap data through 1991 confirm that no effect of ELF on developmental patterns occurred: in two species of velvet mites, seasonal appearance of larvae, deutonymphs and adults has essentially remained unchanged since 1985.

We continue to document seasonal structure of earthworm populations, although the parameter has not yet been subjected to rigorous analysis. These data yield useful background information; for Lumbricus rubellus, for instance, we can now show that approximately 2 to 2.5 years are needed to develop from hatchling to adult. In Aporrectodea tuberculata, we detected yearly fluctuations in immature proportions which were related to reproductive patterns, as influenced by moisture conditions as well as EM fields.

c. Behavioral traits: Weekly trap-catches of arthropods are used to describe the degree of "tracking" of seasonal activity patterns in Test and Control sites. Among approximately 60 species obtained by this method, a dozen are trapped in large enough numbers for

analysis. Activity patterns, which can be strongly seasonal and depend partly on environmental variables (particularly in Collembola) and partly on life cycle events (important in carabid beetles and velvet mites) have not been detectably affected by ELF operation.

In earthworm species of interest to project goals (Aporrectodea tuberculata and Lumbricus rubellus), vertical distribution patterns in response to moisture and temperature have not been affected by antenna operation. Indeed, it is now clear that low coefficients of determination (i.e., a low degree of relationship between moisture and earthworm distribution in the soil profile) indicate a low incidence of moisture stress.

d. Reproduction: Earthworm reproduction (specifically in Aporrectodea tuberculata) has emerged as the group of variables most amenable to analysis as well as to continued experimentation. Tentative conclusions arrived at in previous years have now been validated, at least in part, and further confirmation will be sought during 1993, the last year of field study. Antenna operation does seem to affect reproductive activity of A. tuberculata, but in ways that are both subtle and more complex than originally surmised. Our current conclusions are based on the following evidence:

i. When years were ranked according to average soil moisture during the period critical for earthworm reproduction (June through mid-August), 1990 occupied the top rank. Furthermore, three of the four operational years fell into the top four ranks; i.e., moisture

conditions were relatively propitious during operational years. However, seasonally unmodulated and unusually low numbers of clitellates were observed throughout 1990 (the second year of antenna operation). In 1991-92, some seasonal modulation of clitellate abundance began to re-appear, but numbers still remained below the species' reproductive potential (by comparison with 1984 and 1987).

ii. Multiple regression revealed that soil moisture, after antenna activation, no longer explained variations in clitellate and cocoon abundance; i.e., the normal response of A. tuberculata to moisture fluctuations was altered by an additional factor, which we believe to be EM fields.

iii. Tight correlations between reproductive parameters in A. tuberculata and its sister species in Control, A. turgida, were established for pre-operational years. BACI analyses showed that A. tuberculata reproduction was significantly reduced with respect to A. turgida following antenna activation.

iv. Data for A. tuberculata alone, as well as comparisons with A. turgida, indicated that rates of cocoon production were higher during 1989-92 than during 1984-88. These results are not readily quantified, however, because field-derived data are afflicted with high variances which result in low significance levels. By isolating groups of A. tuberculata in retrievable mesh bags, greater control over experimental conditions was attained, allowing more definitive conclusions (ref. points v. and vi. below).

v. When newly exposed to EM fields (incubation in mesh bags in the Test site), A. tuberculata reproduced at higher rates than when

incubated in Control. A single parameter (cocoon/clitellate ratios, or cocoon production rates) proved to be significantly affected by EM fields. While numbers of clitellates were approximately equal in Test and Control mesh bags, Test adults deposited a greater number of cocoons per adult per unit time.

vi. There is some evidence that, after removal from EM field influence (collected in Test and incubated in mesh bags in Control), an increased number of A. tuberculata became reproductive. The evidence is not statistically tight at this time, and will be tested further during 1993.

Apparent contradictions between field population and experimental (isolation in mesh bags) data can be explained if increased cocoon production rates are viewed as the primary effect of exposure to EM fields. Higher rates imply greater expenditure of energy (in the form of cocoons). It is plausible that adults leave the pool of reproductive individuals sooner (i.e., become post-reproductive after a shorter time period) than they would without EM exposure; the total number of clitellates present in the population at any given time would thereby be reduced. The data also indicate that effects on cocoon production are immediate as well as long-lasting, but that secondary effects on numbers of clitellate adults exhibit a time delay. We believe that continued monitoring of field-incubated earthworms, which are now entering their second year of EM exposure, will clarify current results.

I. SUMMARY OF RESULTS TO DATE

1. Status of data base

a. Soil and litter Arthropoda: All faunal material from the 1991 season has been sorted and catalogued, including developmental stage identifications for selected species. Pertinent analyses are listed below (Table 1) and are discussed in following sections. Data for 1992 will become available in late 1993 to early 1994.

b. Surface-active Arthropoda: Specimens obtained by pit-trapping in 1991 (the last year in which this work element was pursued) have been identified, and summary results are given in this report. Selected data will be presented in greater detail in future refereed publications.

c. Earthworms: Detailed data pertaining to lumbricid field populations, as well as those stemming from isolation experiments (wormbags) are available through 1992. However, only data directly related to ELF project goals are presented in this report.

d. Litter inputs and decomposition: Litterfall estimates are available for the 1992 season (the last year for monitoring this parameter). Litter turnover estimates, based on ash-free dry mass of confined leaves, are not completed at this time. Samples were processed, ground and stored; ashing, however, was delayed by equipment failure.

2. Statistical methods and interpretation

Where possible, we have used BACI (Before and After Control and Impact) tests of [Control - Test] differences for all variables for which pre-ELF and operational data were available. If data could not be normalized, non-parametric Kruskal-Wallis Anova was performed.

For pit-trap data, which are serially correlated over time, ratios of numbers trapped $[(N \text{ on date}_i) / (N \text{ on date}_{i-1})]$ in each site were subjected to BACI tests.

For variables unique to the Test site (phenology of earthworms), multiple regression was the main method of analysis, data being divided into three sets (1984-86, 1987-88, and 1989-92).

The protocol for earthworm isolation experiments was changed in 1992, such that auto- or serial correlation problems were alleviated. Results of tests of normality and homogeneity of variances determined appropriate statistical procedures. In most cases, ANOVA was possible, with Kruskal-Wallis tests as second choice if normality could not be achieved.

Whenever single-date or -year comparisons were of interest, t-tests or Lohrding's q-tests (given equal coefficients of variation) were used.

Interpretation of results was graded into three categories:

- No detectable effect: either absolute values, or the magnitude of between-site differences, did not change following antenna activation;

- ELF effects unlikely: indicates statistical significance, but

examination of raw data showed variables other than EM fields to be equally likely causative agents for observed differences;

Probable ELF effect: evidence for influence of EM fields appeared strong, particularly where effects were detected in two or more related variables or by means of more than one statistical procedure or study technique.

3. Summary of results

Results to date are listed in Table 1 (consistent with Table 1 of our 1991 report, although somewhat expanded). Details relevant to findings and interpretation can be found in the body of this report, which places emphasis on presentation and discussion of variables for which "probable ELF effects" were detected.

Table 1. Summary of results to date, Tasks 5.3. and 5.4. (Arthropoda and Earthworms).

VARIABLE	TEST PROCEDURE	FINDINGS THROUGH (YEAR)	CV ^{a)}	DETECT. LIMIT ^{b)}	AVAIL. PRE-OPER.	PROPOSED OPERATIONAL
SOIL/LITTER ARTHROPODA						
Seasonal abundance (soil):						
Collembola:						
I. minor	BACI	No detectable effect (91)	531.6	250.4	86-88	89-92
I. notabilis	BACI	Unlikely ELF effect (91)	268.1	120.9	86-88	89-92
T. granulata	BACI	No detectable effect (91)	105.8	49.0	86-88	89-92
T. mala	BACI	No detectable effect (91)	49.7	20.4	86-88	89-92
Acarina:						
Mesostigmatid sp. A	K-W ^{c)}	No detectable effect (91)	305.6	-	86-88	89-92
Seasonal abundance (litter):						
Collembola:						
I. notabilis	K-W	No detectable effect (91)	106.8	-	84-88	89-92
O. hexfasciata	K-W	Unlikely ELF effect (91)	173.5	-	84-88	89-92
S. henshawi	K-W	Unlikely ELF effect (91)	138.4	-	84-88	89-92
T. flavescens	K-W	No detectable effect (91)	113.5	-	84-88	89-92
Acarina:						
A. aphidioides	K-W	No detectable effect (91)	325.3	-	84-88	89-92
Nanorchestes sp. A	K-W	No detectable effect (91)	882.8	-	84-88	89-92

a) Coefficient of variation = (SD / mean) x 100, based on pre-operational years

b) Detection limit = $\pm (SD_{\text{pooled}} \times t_{0.05, n1+n2-2}) / (\text{pre-ELF mean}) \times 100$; $SD_{\text{pooled}} = \sqrt{(S_1^2/n_1 + S_2^2/n_2)}$

where 1 = pre-operational, 2 = operational.

c) K-W = Kruskal-Wallis non-parametric test.

Table 1. cont'd:

VARIABLE	TEST PROCEDURE	FINDINGS THROUGH (YEAR)	CV	DETECT. LIMIT	AVAIL. PRE-OPER.	PROPOSED OPERATIONAL
SOIL/LITTER ARTHROPODA						
Population structure:						
I. <u>notabilis</u> : Instar I	BACI	No detectable effect (91)	1054.6	573.5	84-88	89-92
Adults	BACI	No detectable effect (91)	3522.6	2200.4	84-88	89-92
Mesostigmatid sp. A	K-W	No detectable effect for any of 4 stages (91)	196.1 ^a 2225.6 ^a	- -	86-88	89-92
A. <u>aphidoides</u>	K-W	No detectable effect for any of 4 stages (91)	364.4 ^a 678.9 ^a	- -	84-88	89-92
Collembola:						
Community diversity	BACI	No detectable effect (91)	53.6	25.3	86-88	89-92
Community equitability	BACI	Unlikely ELF effect (91)	57.8	25.7	86-88	89-92
SURFACE-ACTIVE ARTHROPODA						
Community diversity:						
Collembola nocturnal	BACI	No detectable effect (91)	777.6	190.3	85-88	89-91
Collembola diurnal	BACI	Unlikely ELF effect (91)	320.7	98.0	85-88	89-91
Total Collembola	BACI	No detectable effect (91)	302.9	83.9	85-88	89-91
Carabidae nocturnal	BACI	No detectable effect (91)	166.0	48.6	85-88	89-91
Carabidae diurnal	BACI	Unlikely ELF effect (91)	394.3	153.8	85-88	89-91
Total Carabidae	BACI	Unlikely ELF effect (91)	189.2	58.4	85-88	89-91
Community equitability:						
Collembola nocturnal	BACI	No detectable effect (91)	214.0	54.5	85-88	89-91
Collembola diurnal	BACI	No detectable effect (91)	665.5	188.7	85-88	89-91
Total Collembola	BACI	No detectable effect (91)	327.7	90.5	85-88	89-91
Carabidae nocturnal	BACI	No detectable effect (91)	339.7	106.1	85-88	89-91
Carabidae diurnal	BACI	Unlikely ELF effect (91)	231.1	82.2	85-88	89-91
Total Carabidae	BACI	No detectable effect (91)	316.6	77.5	85-88	89-91

a) Minimum and maximum CV observed among 4 developmental stages

Table 1. cont'd:

VARIABLE	TEST PROCEDURE	FINDINGS THROUGH (YEAR)	CV	DETECT. LIMIT	AVAIL. PRE-OPER.	PROPOSED OPERATIONAL
SURFACE-ACTIVE ARTHROPODA						
Seasonal activity patterns:						
Collembola:						
<u>S. henschawi</u>	BACI	No detectable effect (91)	1164.7	349.5	85-88	89-91
<u>S. lepus</u>	BACI	No detectable effect (91)	3621.8	1156.4	"	"
<u>O. hexfasciata</u>	BACI	No detectable effect (91)	1339.5	330.1	"	"
<u>T. flavescens</u>	BACI	No detectable effect (91)	687.7	263.0	"	"
Acarina:						
<u>Abrolophus</u> sp.	BACI	No detectable effect (91)	221.8	5294.6	"	"
<u>T. auroraense</u>	BACI	No detectable effect (91)	577.1	205.3	"	"
<u>Nanorchestes</u> sp. A	BACI	No detectable effect (91)	574.2	159.2	"	"
Carabidae:						
<u>P. melanarius</u>	BACI	No detectable effect (91)	890.7	337.0	"	"
<u>P. pensylvanicus</u>	BACI	No detectable effect (91)	595.7	251.7	"	"
<u>P. coracinus</u>	BACI	No detectable effect (91)	829.3	263.9	"	"
<u>S. impunctatus</u>	BACI	No detectable effect (91)	7316.2	3980.8	"	"
<u>H. fuliginosus</u>	BACI	No detectable effect (91)	769.9	338.2	"	"

Table 1. cont'd:

VARIABLE	TEST PROCEDURE	FINDINGS THROUGH (YEAR)	CV	DETECT LIMIT	AVAIL. PRE-OPER.	PROPOSED OPERAT.
EARTHWORMS						
Community diversity (TEST)	t-tests	No detectable effect (92)	10.0 ± 1.7 ^{b)}	-	84-88	89-93
A. tuberculata vs. turgida						
Clitellate density	BACI	Probable ELF effect (92)	669.2	246.4	84-88	89-93
Proportion clitellate	BACI	Probable ELF effect (92)	113.0	37.9	84-88	89-93
Cocoon abundance	BACI	Probable ELF effect (92)	520.1	231.3	84-88	89-93
A. tuberculata (TEST)						
Vertical distribution	MR ^{a)}	No detectable effect (92)	-	-	84-88	89-93
Proportion clitellate	MR	Probable ELF effect (92)	-	-	84-88	89-93
Clitellate density	MR	Probable ELF effect (92)	-	-	84-88	89-93
Cocoon density	MR	Probable ELF effect (92)	-	-	84-88	89-93
L. rubellus (TEST)						
Vertical distribution	MR	No detectable effect (92)	-	-	84-88	89-93
Reproduction	MR	No detectable effect (92)	-	-	84-88	89-93
D. octaedra						
Vertical distribution	MR	No detectable effect (91)	-	-	84-88	89-93
Clitellate density	MR	No detectable effect (91)	-	-	84-88	89-93
Cocoon density	MR	No detectable effect (91)	-	-	84-88	89-93

a) MR = multiple regression; see text for details

b) Mean CV ± SD for 5 pre-operational years

Table 1. cont'd:

VARIABLE	TEST PROCEDURE	FINDINGS THROUGH (YEAR)	CV	DETECT. LIMIT	AVAIL. PRE-OPER.	PROPOSED OPERAT.
EARTHWORMS						
A. tuberculata isolation exp. (1992 data):						
TEST PROVENANCE						
Body mass	ANOVA	Probable ELF effect (92)	26.7, 24.7 a)	-	-	91-93
Cocoon mass	ANOVA	No detectable effect (92)	19.0, 27.7 a)	-	-	91-93
Proportion clitellate	ANOVA	Probable ELF effect (92)	26.0, 17.9 a)	-	-	91-93
Cocoon production rate	ANOVA	Probable ELF effect (92)	(see text)	-	-	91-93
FIRE TOWER PROVENANCE						
Body mass	t- or q-tests	Probable ELF effect (92)	23.5, 23.3 a)	-	-	91-93
Cocoon mass	ANOVA	No detectable effect (92)	21.9, 29.3 a)	-	-	91-93
Proportion clitellate	K-W	No detectable effect (92)	54.9, 59.6 a)	-	-	91-93
Cocoon production rate	ANOVA	Probable ELF effect (92)	(see text)	-	-	91-93
LITTER INPUTS AND DECOMPOSITION						
Seasonal litter inputs:						
Maple	BACI	No detectable effect (92)	712.5	322.2	84-88	89-92
Basswood	BACI	No detectable effect (92)	2464.3	1003.4	84-88	89-92
Total	BACI	No detectable effect (92)	344.8	96.4	84-88	89-92
Seasonal litter standing crops:						
Oven-dry mass	BACI	No detectable effect (91)	92.2	36.1	85-88	89-91
Ash-free dry mass	BACI	No detectable effect (91)	58.7	42.7	87-88	89-91
Decomposition:						
Mass loss (AFDW)	ANOVA	No detectable effect (91)	180.1	54.6	86,89	90-93

a) CV for Test, Control respectively

II. ENVIRONMENTAL MONITORING

1. Precipitation

In 1992, rainfall deficits occurred mainly in May and early June (Fig. 1). Total precipitation in July, August and September was close to 30-year averages (Table 2); totals for October are not shown in Table 2, because raingauges were dismantled on October 19 after snowfall had occurred.

Table 2. Monthly precipitation totals in Test and Control, 1992, and 30-year means for the area at large (Crystal Falls Weather Station).

	May	June	July	Aug	Sep	Total
Control	38.8	43.4	83.4	103.9	86.2	355.7
Test	54.6	26.6	108.1	88.3	97.9	375.5
30-yr mean	81.0	105.4	91.4	98.5	84.6	460.9

2. Soil and litter moisture

Much as in 1991, soil and litter moisture estimates tracked each other well in 1992 (Fig. 2), A horizon moisture falling below 20% in late May and early June, during a period of low rainfall.

Although we attempted to use TDR (Time Domain Reflectometry) sensors for monitoring soil moisture, we were unable to obtain satisfactory results for "gravimetric vs. TDR" regressions for A horizon samples. Small-scale heterogeneity in texture and root content of these forest soils forces us to continue using gravimetric moisture determinations.

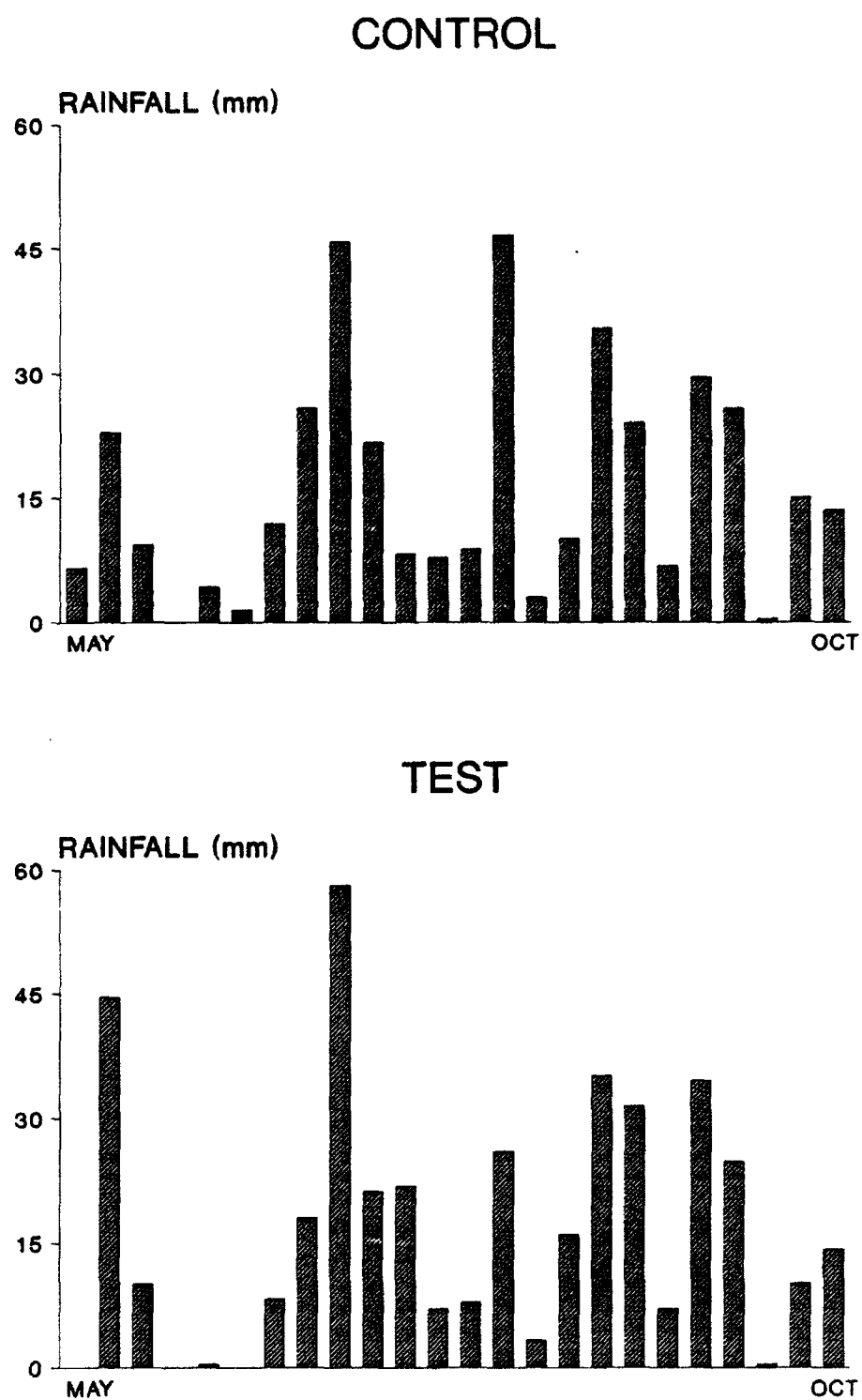


Fig. 1. Total weekly precipitation in Test and Control, 1992.

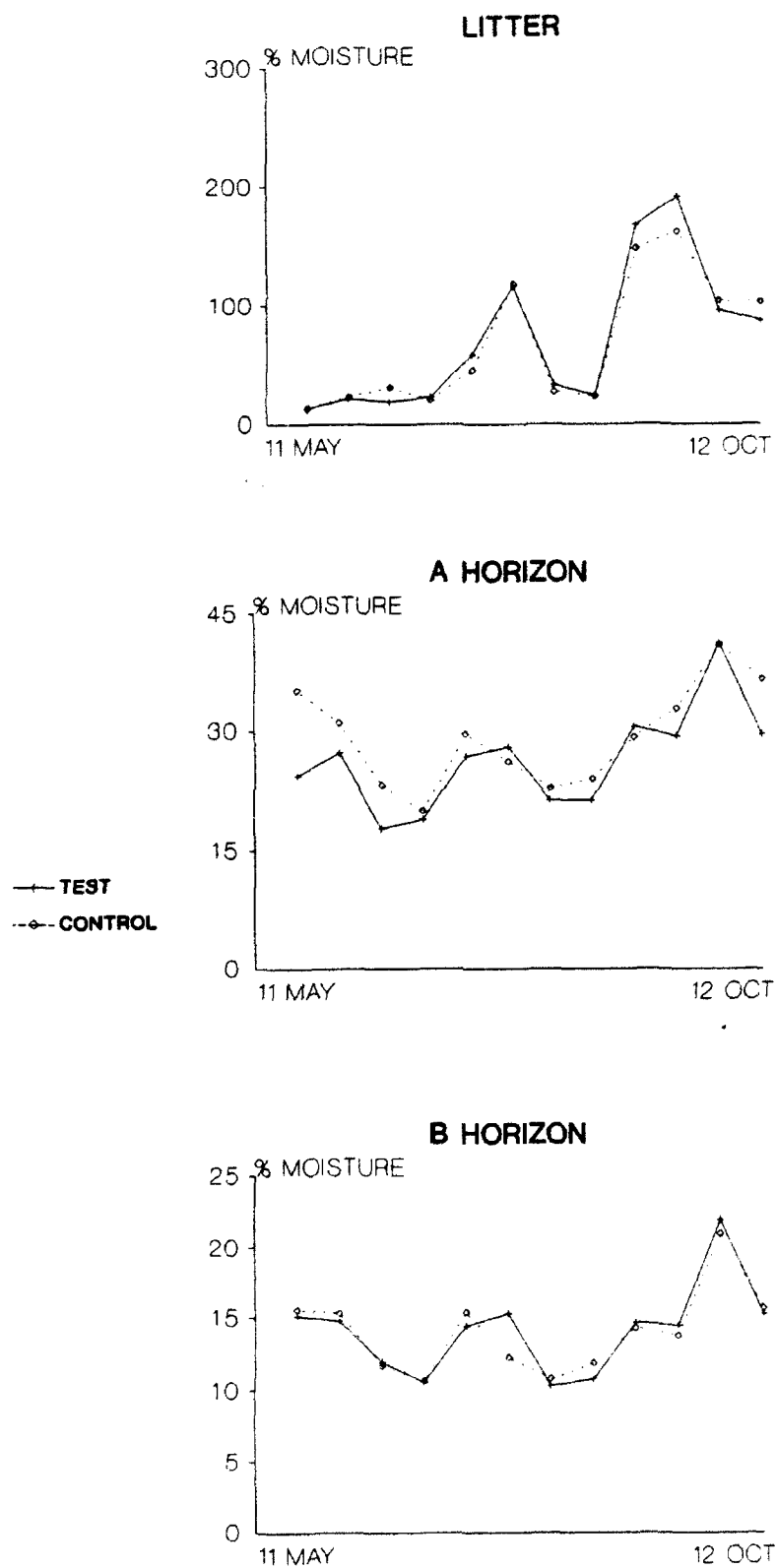


Fig. 2. Litter and soil moisture (% gravimetric), 1992.

Moisture of the A horizon is a major variable used for earthworm data analyses. In order to characterize yearly variations in edaphic conditions impinging on lumbricids, average moisture during the time period critical for reproductive performance was calculated (Table 3). In all years, that period clearly extended over sampling dates 3 through 8 (approximately early June through mid-August).

Ranked according to average moisture, 1990 and 1987 emerged as the most propitious years, followed by 1989 and 1991. The top four ranks thus included three of the four operational years. As a further means of characterizing potential stress conditions for lumbricids, the number of dates when moisture fell below 20%, and the number of consecutive dates with low moisture, are also listed in Table 3. Together with ranges, these data identify edaphic conditions relatively well. In 1991, for instance, a minimum value of 13.4% represented a single occurrence of moisture deficit. In mid-season of 1988, on the other hand, moisture was low on five of six dates, and maximum and minimum values were the lowest ever recorded (Table 3).

3. Temperature

Average weekly air temperatures in Test and Control are shown in Fig. 3. The 1992 season was generally cooler than previous seasons, which is illustrated in Fig. 4 by means of a comparison between 1991 and 1992. With respect to soil temperatures, we have been plagued with equipment failure throughout the year. By means of YSI telethermometer measurements (and comparison to datalogger recordings) we determined that electronic sensors in the Test site were reliable, but not those

in Control. However, records from past years have shown that Test and Control data are essentially interchangeable. We have replaced missing data (approx. 20% of total records) via regression analyses on air temperature, and intend to replace faulty equipment before the 1993 season begins.

Mean weekly temperatures obtained at 5 cm depth in Test in 1991 and 1992 (Fig. 5) again illustrate the relative coolness of the latter season.

Table 3. Mean percent A horizon moisture (sampling dates 3 to 8), range of single-date estimates, and number of occasions on which moisture fell below 20%.

YEAR	RANK	MEAN %	SD	N DATES <20%	N CONSECUT. DATES <20%	RANGE
1984	5	23.04	3.30	1	0	16.6 - 25.6
1985	7	20.78	2.17	3	3	15.8 - 26.7
1986	8	19.76	1.67	3	2	16.6 - 24.1
1987	2	25.29	1.78	0	0	21.4 - 29.8
1988	9	14.60	2.26	5	5	8.1 - 20.5
1989	3	24.97	1.51	0	0	20.0 - 27.4
1990	1	25.46	2.20	0	0	20.0 - 33.3
1991	4	23.45	2.41	1	0	13.4 - 30.5
1992	6	22.37	2.04	2	2	17.8 - 28.1

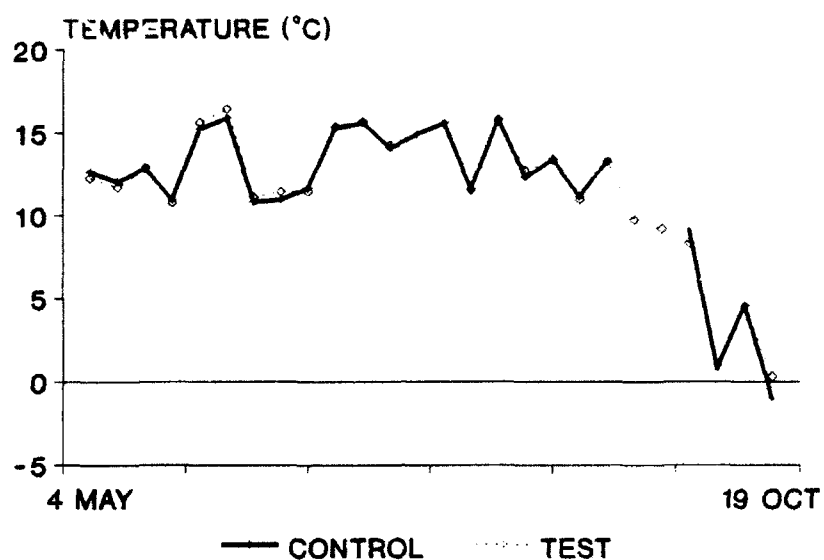


Fig. 3. Average weekly air temperatures in Test and Control, 1992.

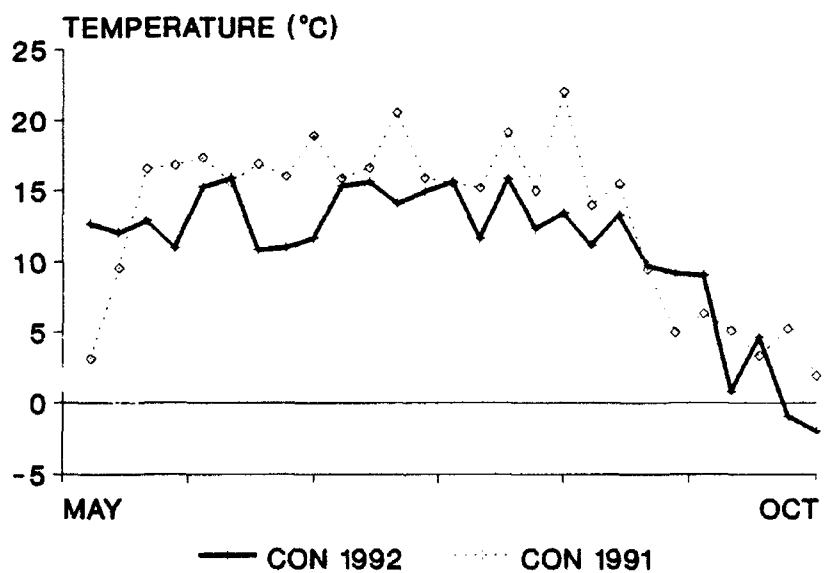


Fig. 4. Average weekly air temperatures in Control, 1991 vs. 1992.

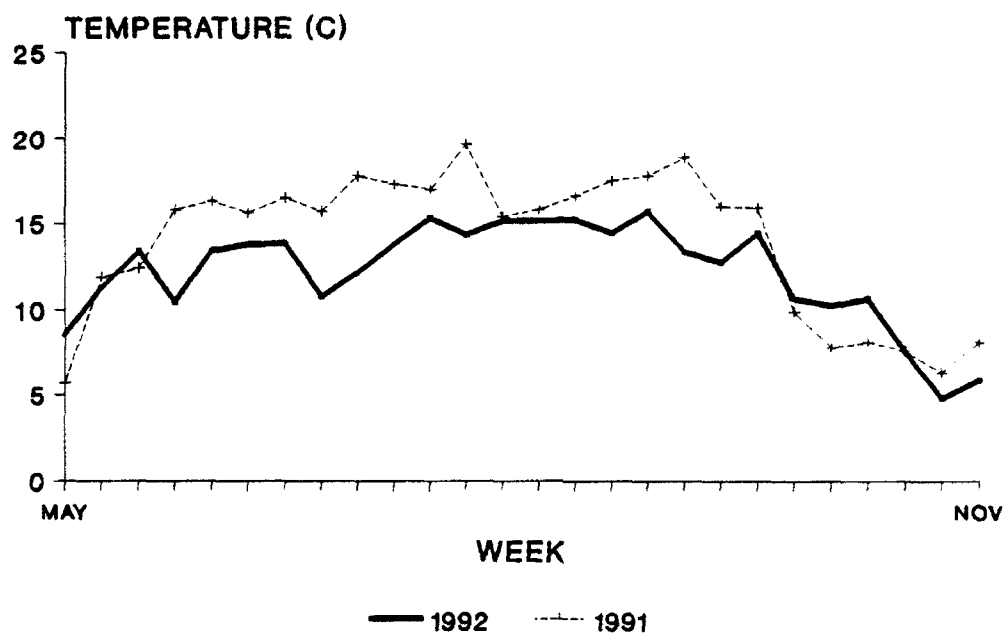


Fig. 5. Weekly mean temperature of the A horizon in the Test site at 5 cm depth, 1991 and 1992.

III. SOIL AND LITTER ARTHROPODA

1. Collembola:

Mean annual densities of the most abundant species, as well as family totals, provide an overview of long-term population fluctuations in Control and Test sites (Table 4).

Most species are recovered from both litter and soil strata. Analyses of seasonal densities were performed for the stratum in which a given species occurs most frequently in both sites.

Statistical results for three species (BACI or Kruskal-Wallis tests, Table 1) were significant when pre-operational data were compared to operational years. Equitability of the collembolan communities also diverged in 1989-91; however, equitability of the Test community has remained relatively unchanged ($P= 0.09$), while that of the Control community has experienced an increase ($P= 0.01$).

We suggest strongly that long-term shifts in abundance of community components, unrelated to ELF activation, underlie these results. Drastic year-to-year density fluctuations of total Hypogastruridae and Onychiuridae in Control, or of single species in both sites, appear to be the rule (Table 4).

We present a few simple graphs to illustrate the basis for our conclusions. Abundance of Collembola in Control, typically more than twice that in Test (Fig. 6), was much reduced in 1991 when compared to the previous 5 years (years for which extraction efficiency data are available). To a great degree, this reduction was due to lower onychiurid numbers (Fig. 7).

Table 4. Mean annual density (litter + soil) of selected species and family totals of Collembola in Test and Control; years without extraction efficiency data (1984 and 1985) not included.

	1986		1987		1988		1989		1990		1991	
	T	C	T	C	T	C	T	C	T	C	T	C
<u>S. henshawi</u>	224	357	198	321	356	325	180	201	305	240	187	148
<u>SMINTHURIDAE</u>	299	490	418	471	453	400	311	319	423	324	274	203
<u>I. notabilis</u>	1782	2690	2220	3739	1076	1542	1661	2210	2383	2138	1749	1175
<u>I. minor</u>	408	292	304	541	137	200	393	224	350	323	415	383
<u>ISOTOMIDAE</u>	2586	3571	3099	4996	1400	2175	2219	3271	3031	3473	2542	2386
<u>T. flavescens</u>	495	59	876	24	277	142	175	12	246	37	206	11
<u>O. hexfasciata</u>	234	28	528	73	170	95	42	70	124	101	197	25
<u>E. comparata</u>	84	34	128	58	72	222	46	255	39	265	25	67
<u>ENTOMOBRYIDAE</u>	1416	173	2563	213	1035	514	708	424	945	503	762	156
<u>NEELIDAE</u>	19	293	81	281	13	116	57	148	94	154	134	191
<u>T. mala</u>	2343	17870	4554	24347	4055	17138	5177	17935	1896	13746	3288	8215
<u>T. granulata</u>	3551	5658	5563	11254	4547	7721	4476	6166	3400	7027	4027	4708
<u>T. iowensis</u>	1554	589	2449	4188	2356	2767	1879	6693	2123	7096	2506	2467
<u>T. clavata</u>	746	673	723	1989	321	742	681	535	535	519	519	219
<u>ONYCHIURIDAE</u>	8538	25603	13756	43425	11893	28860	12995	31739	8505	28992	10806	15847
TOTAL COLLEMBOLA	13387	30856	20791	51028	15333	32793	16985	36752	13814	35238	15272	19478
TOTAL N SPECIES	46	55	46	47	45	44	46	48	49	53	51	52

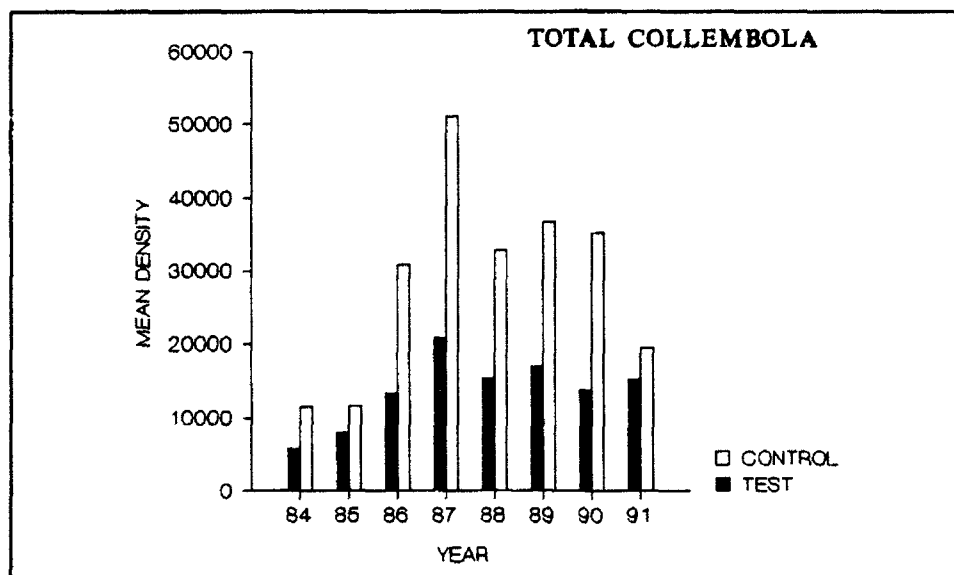


Fig. 6. Annual mean density of total Collembola (1984-85 not corrected for extraction efficiency).

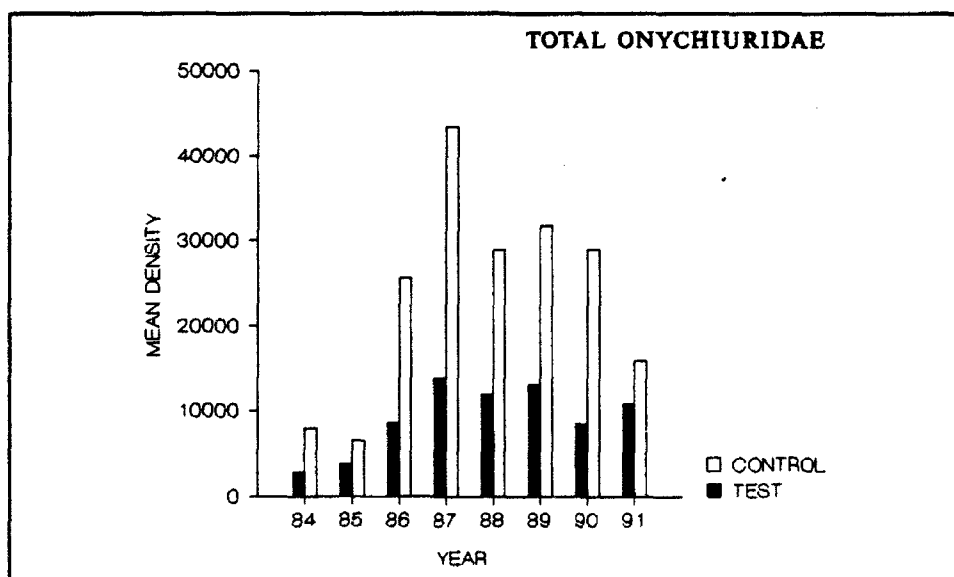


Fig. 7. Annual mean density of Onychiuridae (1984-85 not corrected for extraction efficiency).

At the species level, Test/Control abundance relationships have also changed over time. In the Control site, Isotoma notabilis, Sminthurinus henshawi, and both of the dominant Tullbergia spp have undergone decreases (Figs. 8-12), while roughly opposite trends were observed for Orchesella hexfasciata (Fig. 13). The magnitude of between-site differences recorded prior to 1989 (or prior to 1988 in some cases) was thus changed or even reversed during operational years. We suggest that we are observing long-term fluctuations which produce occasional differences significant in statistical terms, but which would disappear (and re-occur) at random intervals if monitoring were continued over several years.

With respect to population structure of Isotoma notabilis, a significant preponderance of instars I in Control, 1989, now proves to have been a single-year aberrant occurrence much like that observed in 1986 (Fig. 14). With two more years of data at hand, seasonal [Control - Test] differences were no longer significant for either hatchlings or adults (Fig. 15).

2. Acarina:

Analyses of date-specific abundances of total populations, and of seasonal numbers (Mesostigmatid sp. A) or proportions (Asca aphidioides) in each of four developmental stages showed that operational data did not differ from pre-operational data (Table 1). Abundance fluctuations were generally as variable in 1989-91 as they had been in 1984-88 (Figs. 16-18). In 1991, for instance, relative densities of Nanorchestes in Test and Control

leaf litter were reminiscent of those in 1985 and 1988 (Fig. 16); overall abundance fluctuations of the species (Fig. 17) remained well correlated ($R^2 = 0.89$). No evidence of potential ELF effects exists for any of the species monitored.

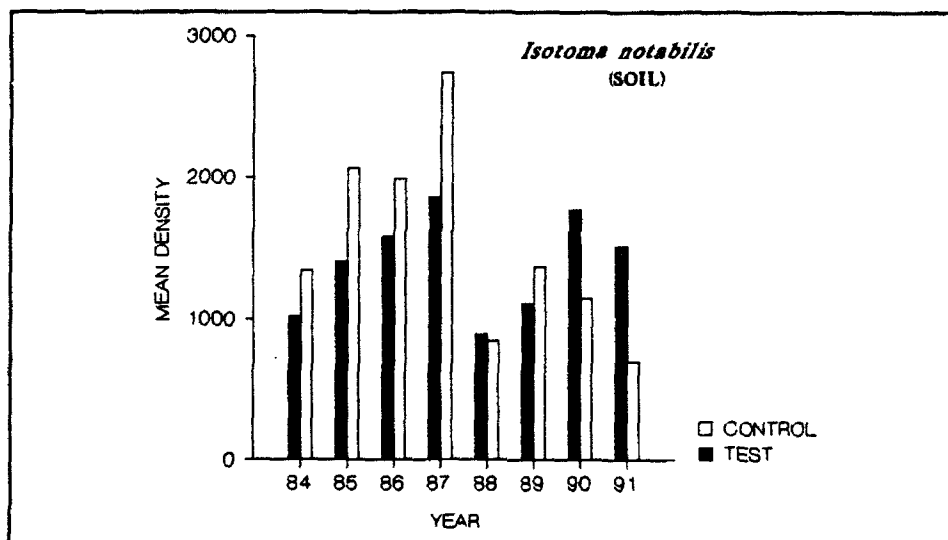


Fig. 8. Mean annual density of *Isotoma notabilis* in Test and Control soil.

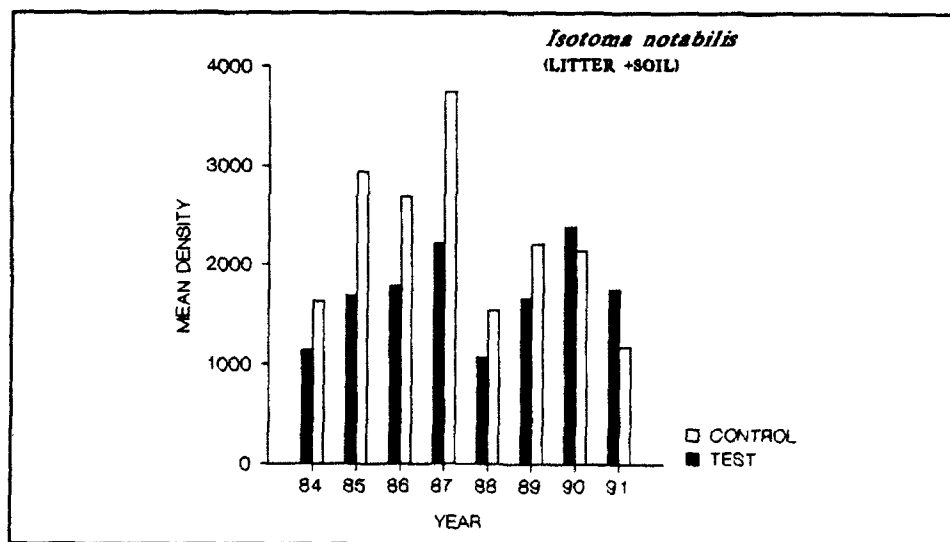


Fig. 9. Mean annual density of *Isotoma notabilis* in Test and Control (litter and soil density estimates summed).

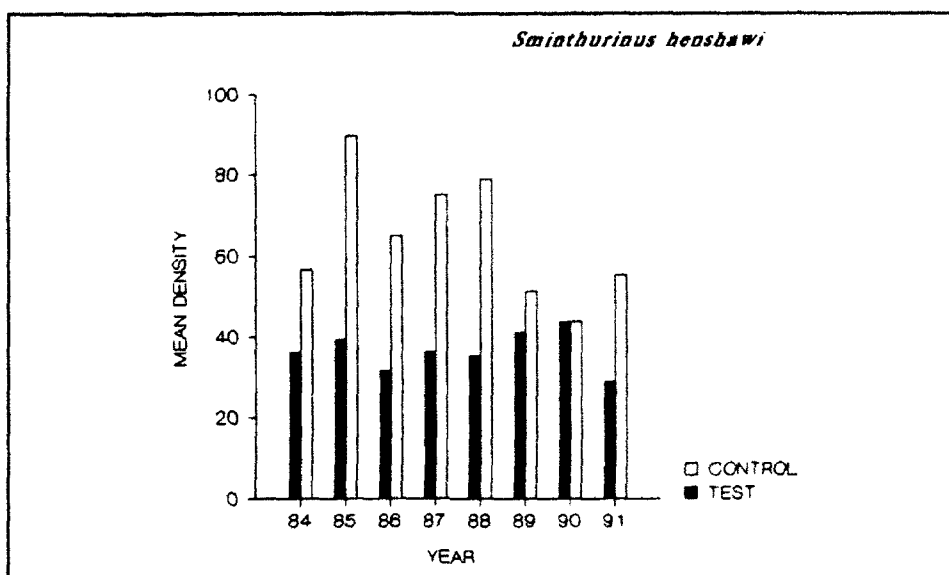


Fig. 10. Mean annual density of *Sminthurinus henshawi* in Test and Control leaf litter.

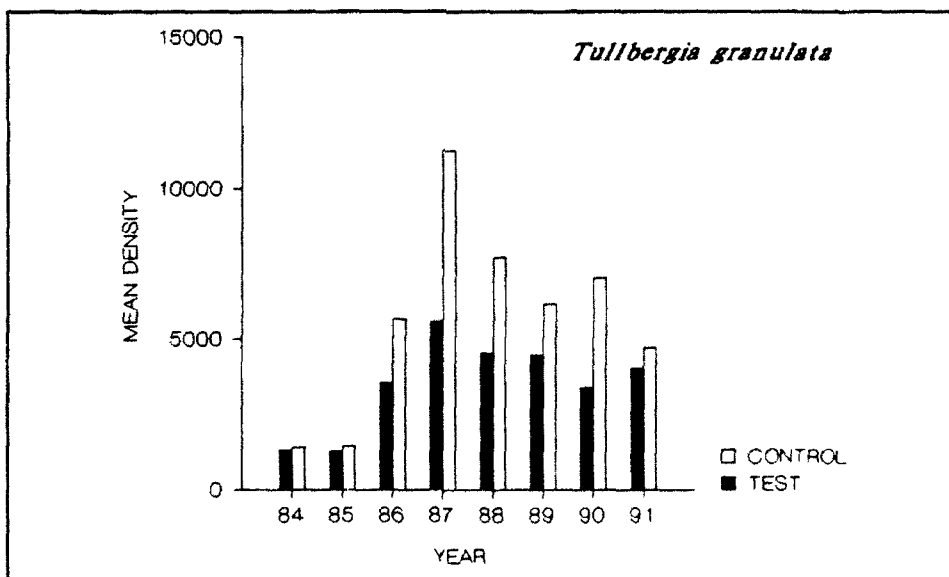


Fig. 11. Mean annual density of *Tullbergia granulata* in Test and Control soil (1984-85 not corrected for extraction efficiency).

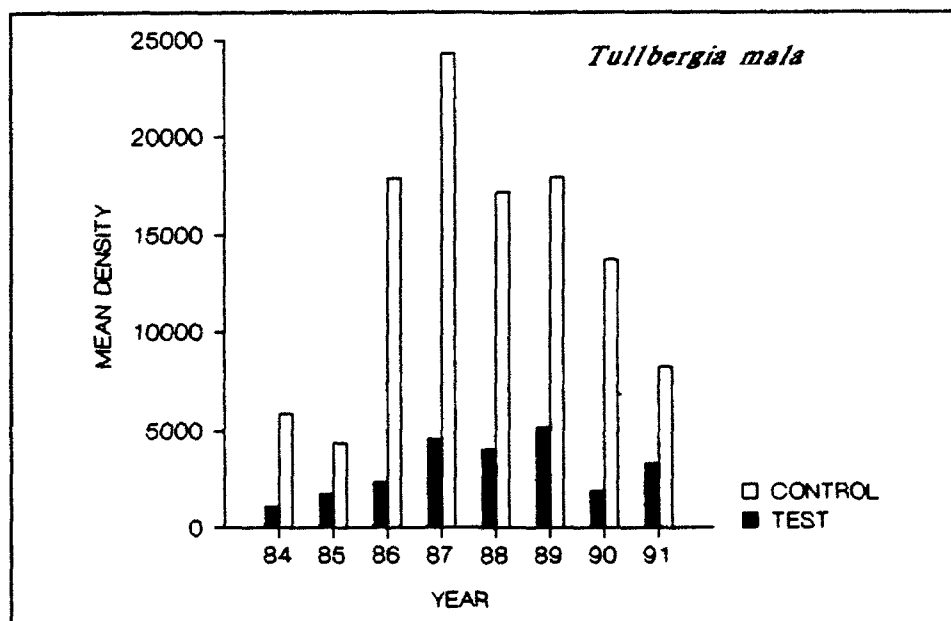


Fig. 12. Mean annual density of *Tullbergia mala* in Test and Control soil (1984-85 not corrected for extraction efficiency).

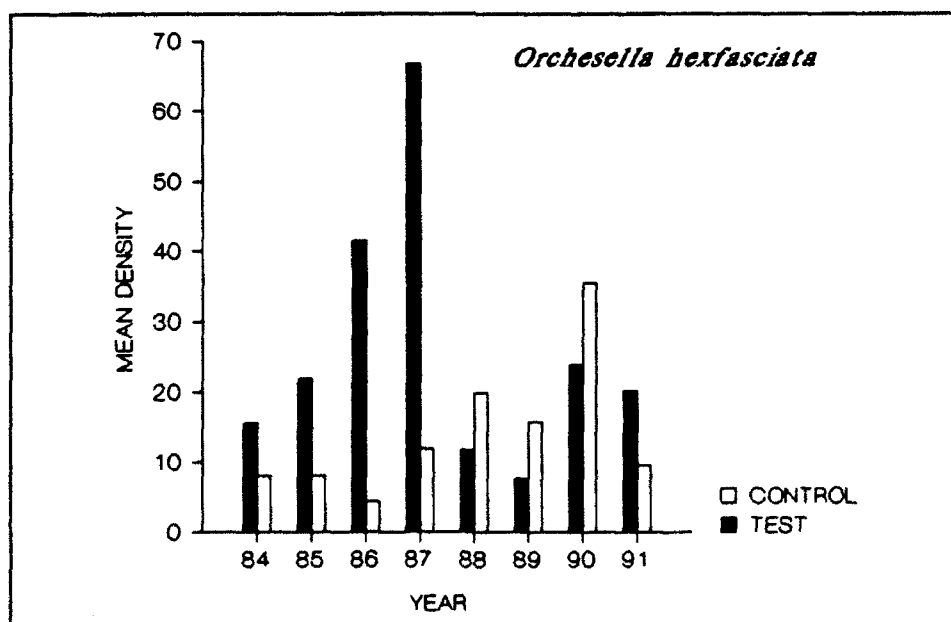


Fig. 13. Mean annual density of *Orchesella hexfasciata* in Test and Control leaf litter.

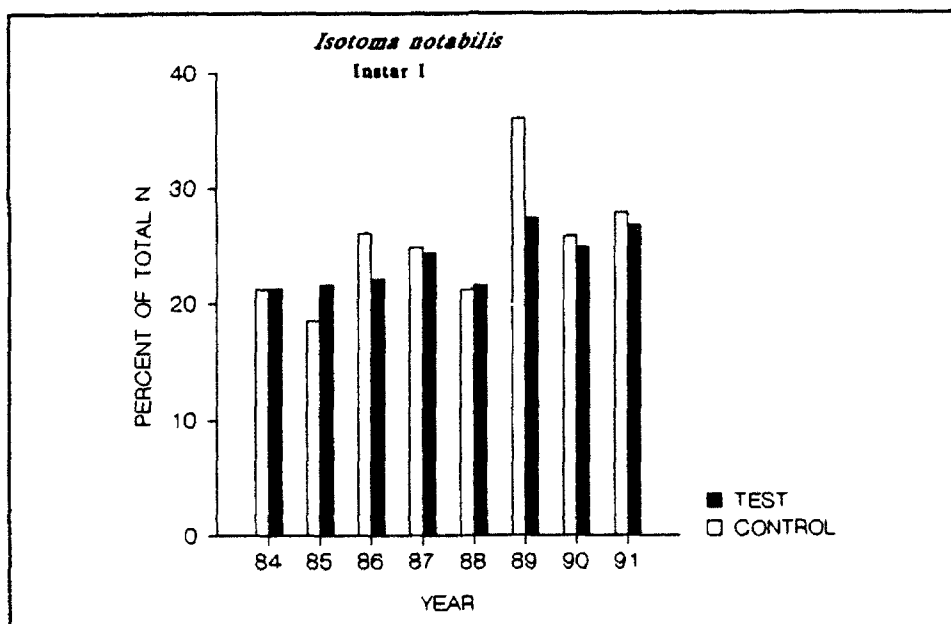


Fig. 14. Frequency of instars I of *Isotoma notabilis* (in percent of total annual population).

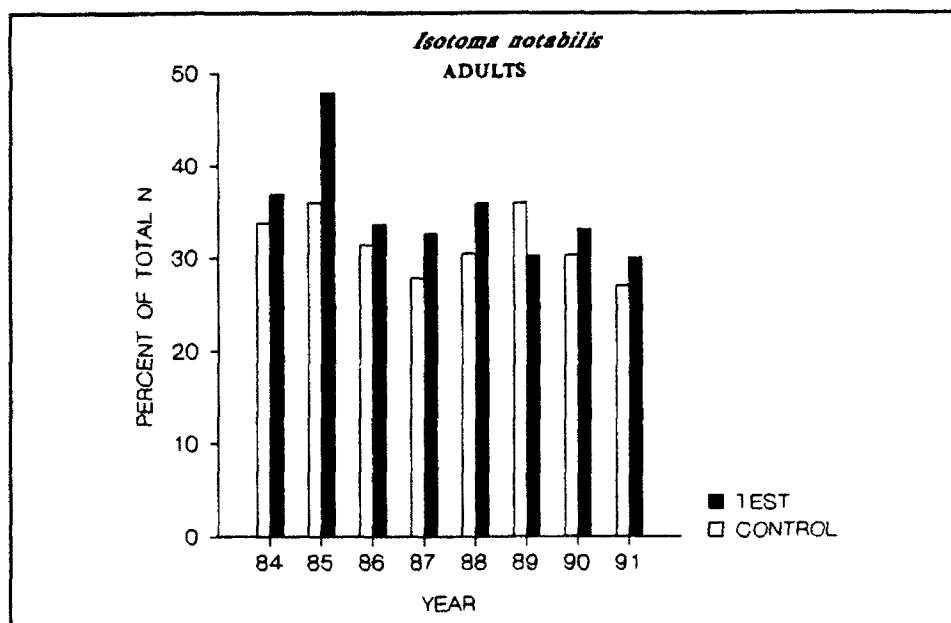


Fig. 15. Frequency of adults of *Isotoma notabilis* (in percent of total annual population).

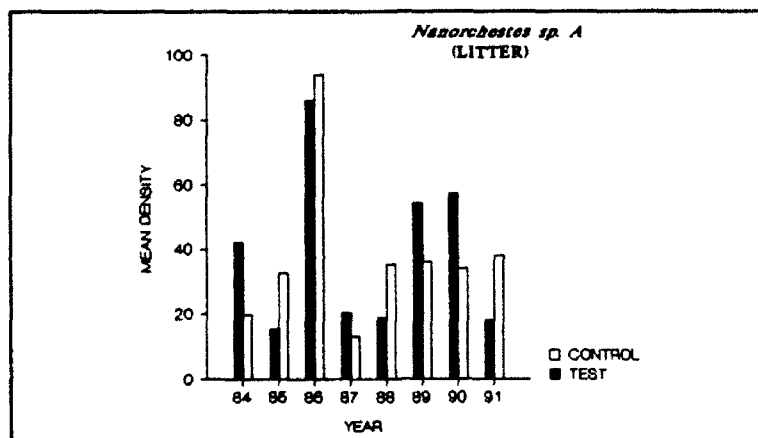


Fig. 16. Mean annual density of *Nanorchestes* sp. A in litter.

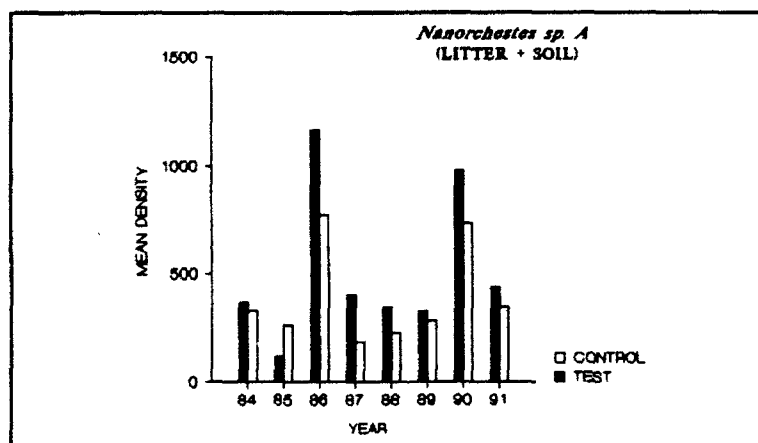


Fig. 17. Mean annual total density of *Nanorchestes* sp. A.

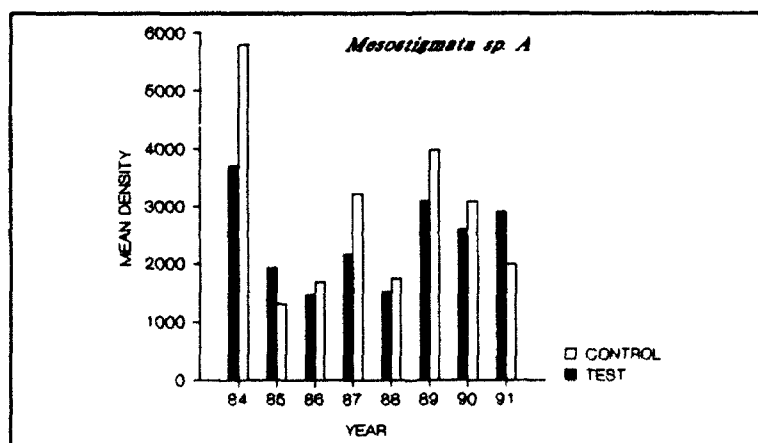


Fig. 18. Mean annual density (soil) of *Mesostigmatid* sp. A.

IV. SURFACE-ACTIVE ARTHROPODA

No detectable responses to ELF activation have occurred with respect to seasonal activity patterns of arthropods. Data through 1991 are now available, and represent the completed data base for this project element. Detailed data are being prepared in manuscript form. In this report, we simply present large-scale summaries, as well as a few species-specific examples of seasonal activity patterns.

1. Collembola:

Day- and night-catches of Collembola, as well as combined totals per date, showed that community diversity increased during the 1989-91 period, particularly in the Test site (Table 4). BACI [Control - Test] analyses of pre- vs. operational periods yielded significant results only for H' of the diurnal assemblage of species. We do not consider this result biologically significant, given the wide year-to-year variation in numbers of various species captured. Examples of this variability can be found in Table 5: e.g., Dicyrtoma aurata in Control, or Tomocerus flavescens and Orchesella hexfasciata in both sites.

Seasonal patterns of activity as reflected in weekly catch totals have remained roughly coincident between sites (Fig. 19 shows data for O. hexfasciata as example). Correlations between Test and Control catches have shown great variability in the past, and have remained variable through 1991. No clear changes attributable to ELF activation have been detected.

Table 5. Diversity and equitability indices (means \pm SD) for surface-active collembolan communities in pre-operational and operational periods. (N dates 1985-88 = 103; N dates 1989-91 = 76).

		TEST			CONTROL			BACI
		1985-88	1989-91	P=	1985-88	1989-91	P=	P=
Diurnal								
	H'	1.59 \pm 0.310	1.74 \pm 0.338	0.03	1.48 \pm 0.027	1.51 \pm 0.352	NS	0.03
	S'/S	0.31 \pm 0.075	0.31 \pm 0.075	NS	0.32 \pm 0.068	0.30 \pm 0.068	NS	NS
Nocturnal								
	H'	1.55 \pm 0.284	1.68 \pm 0.233	0.001	1.50 \pm 0.330	1.64 \pm 0.246	0.004	NS
	S'/S	0.31 \pm 0.069	0.34 \pm 0.076	0.007	0.37 \pm 0.088	0.37 \pm 0.065	NS	NS
Total								
	H'	1.71 \pm 0.287	1.84 \pm 0.270	0.002	1.60 \pm 0.268	1.65 \pm 0.281	NS	NS
	S'/S	0.27 \pm 0.052	0.28 \pm 0.060	NS	0.30 \pm 0.068	0.29 \pm 0.064	NS	NS

Table 6. Total annual pit-trap catches of selected taxa of Collembola, 1985-1991.

	T E S T							C O N T R O L						
	1985	1986	1987	1988	1989	1990	1991	1985	1986	1987	1988	1989	1990	1991
<u>S. henshawi</u>	1637	1435	1992	2811	3065	3196	2364	2606	2934	4123	5084	3675	2666	2475
<u>S. lepus</u>	669	236	1049	503	1438	1375	1335	397	375	1019	824	724	505	1129
<u>D. aurata</u>	5	0	4	1	0	22	1	468	976	2198	448	403	233	145
SMINTHURIDAE	2423	1709	3124	3398	4841	4870	3943	3593	4379	7607	6770	5368	3627	4407
<u>T. flavescens</u>	4213	1965	2429	1684	641	1033	2198	842	242	280	165	237	170	90
<u>O. hexfasciata</u>	3201	3402	4137	3426	738	1767	3434	1099	421	1180	3549	1672	2976	1500
<u>E. comparata</u>	35	80	119	150	57	90	69	287	87	157	1493	440	731	92
<u>E. nivalis</u>	531	1057	294	291	218	326	546	4	14	34	77	104	243	172
<u>L. paradoxus</u>	22	6	37	123	22	229	1377	1142	961	2701	2649	3783	4385	2717
ENTOMOBRYIDAE	8433	7238	8209	7186	2275	3843	8283	3479	1752	4495	8100	6308	8578	4793
HYPOGASTRURIDAE	80	90	191	196	420	269	325	2122	292	456	463	798	964	618
ISOTOMIDAE	582	513	486	292	484	318	550	751	392	562	188	552	297	464
TOTAL ALL SPP.	11518	9550	12010	11072	8020	9301	13101	9946	6815	13120	15522	12964	13467	10282
TOTAL N SPP.	37	29	31	34	37	33	35	33	28	32	32	33	33	36

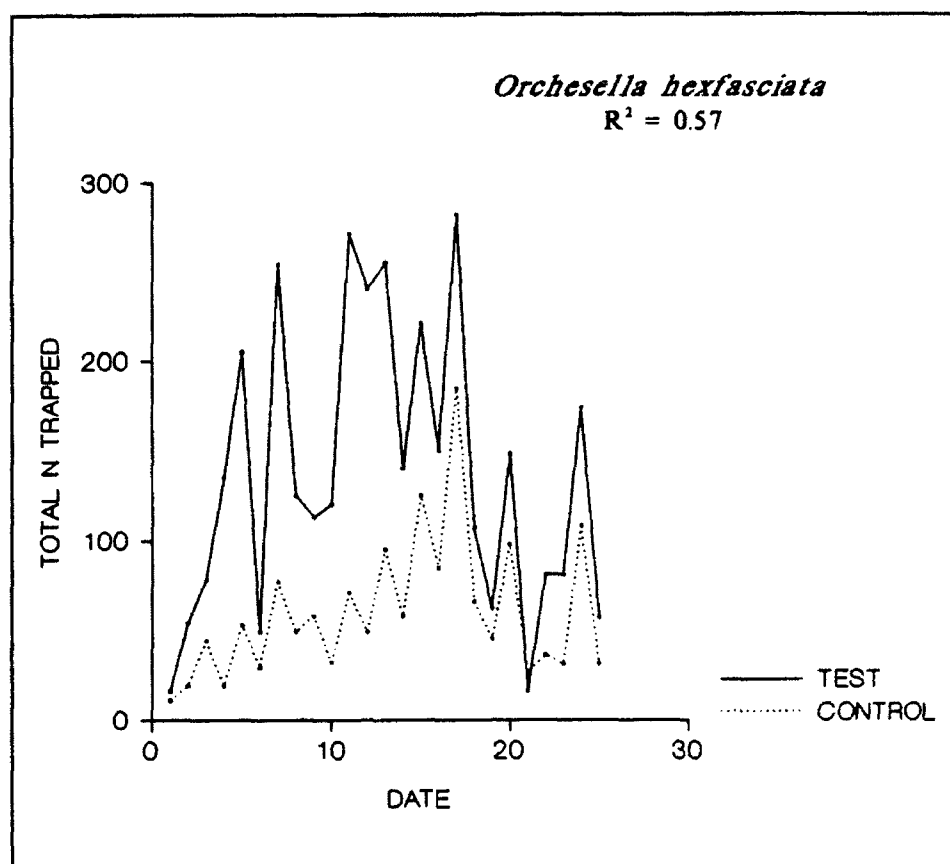


Fig. 19. Weekly catches of *Orchesella hexfasciata*, 1991.

2. Acarina:

Re-examination of velvet mite specimens proved that *Leptus* sp. was captured in numbers too low for analysis. Activity of three other species commonly trapped in both sites (yearly totals are listed in Table 6) showed no detectable effects of antenna activation (Table 1).

Correlations between weekly catches in Test and Control have typically been very tight for *Trombidium auroraense*, but somewhat variable for *Nanorchestes* sp. A and *Abrolophus* sp. (Table 7).

Velvet mites are almost exclusively day-active, unlike Nanorchestes, in which the degree of diurnality has varied between years as well as sites (Figs. 20-22). These tendencies did not change following ELF activation.

Table 7. Total number of three mite species captured during 1985-1989 (night and day catches summed).

YEAR	1985	1986	1987	1988	1989	1990	1991
<u>T. auroraense</u>							
TEST	206	281	202	291	371	318	253
CONTROL	599	731	634	748	724	385	246
<u>Abrolophus</u> sp.							
TEST	309	335	155	387	200	580	267
CONTROL	713	431	226	714	282	293	108
<u>Nanorchestes</u> sp. A							
TEST	1322	4405	1746	2216	1810	3872	2593
CONTROL	808	5926	2329	4984	2697	2370	1350

Table 8. Correlation R^2 for weekly [night + day] catches of Acarina in Test and Control sites.

YEAR	1985	1986	1987	1988	1989	1990	1991
<u>T. auroraense</u>	0.85	0.92	0.86	0.92	0.96	0.92	0.98
<u>Abrolophus</u> sp.	0.94	0.86	0.62	0.72	0.83	0.63	0.57
<u>Nanorchestes</u> sp.	0.72	0.74	0.50	0.85	0.86	0.30	0.61

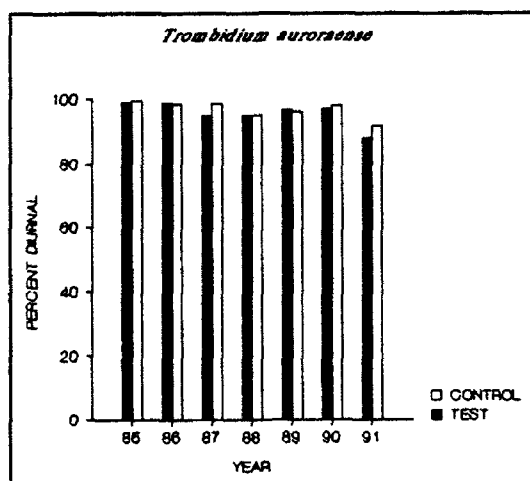


Fig. 20

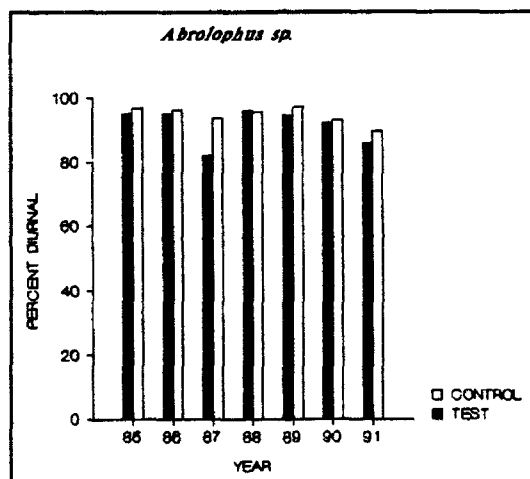


Fig. 21

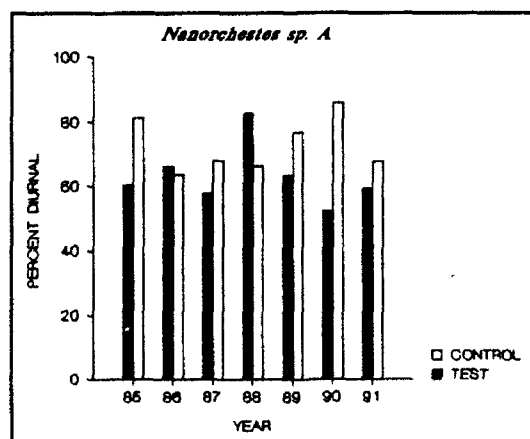


Fig. 22

Figs. 20-22. Percent diurnality of Acarina $[(N \text{ day} / N \text{ total}) \times 100]$.

While catches of Nanorchestes sp. consist entirely of adults (Fig. 23 illustrates 1991 data), those of velvet mites reflect seasonal developmental patterns of the three active stages (larvae, deutonymphs and adults).

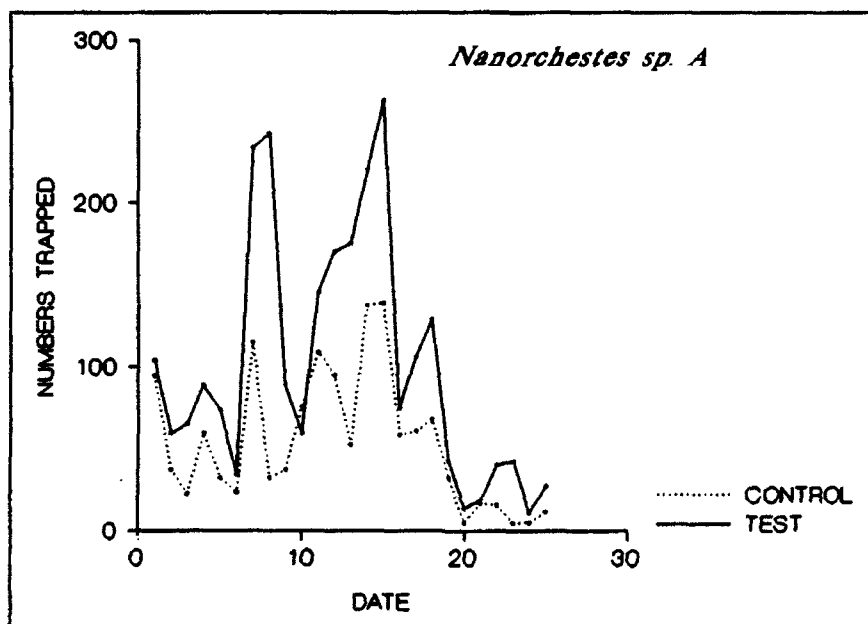


Fig. 23. Weekly catches (May 7 to October 22, 1991) of Nanorchestes sp. A in Test and Control.

Trombidium auroraense overwinter mainly as adults, with deutonymphs as a lesser component of spring-active individuals (Fig. 24). Eggs deposited in May and June give rise to larvae which actively search for insect hosts. Deutonymphs appear in the last third of the season and either overwinter as such or develop to adults in late fall and early spring.

Abrolophus exhibits a distinctly different life cycle. Larvae

appear in early spring, followed by deutonymphs and adults, which are captured with rapidly decreasing frequency until their disappearance in mid-September (Fig. 25 illustrates data from a pre-ELF and an operational year). Be it in terms of numbers or of seasonal distribution of developmental stages (Fig. 26), ELF activation clearly did not affect life cycles or activity patterns of either species.

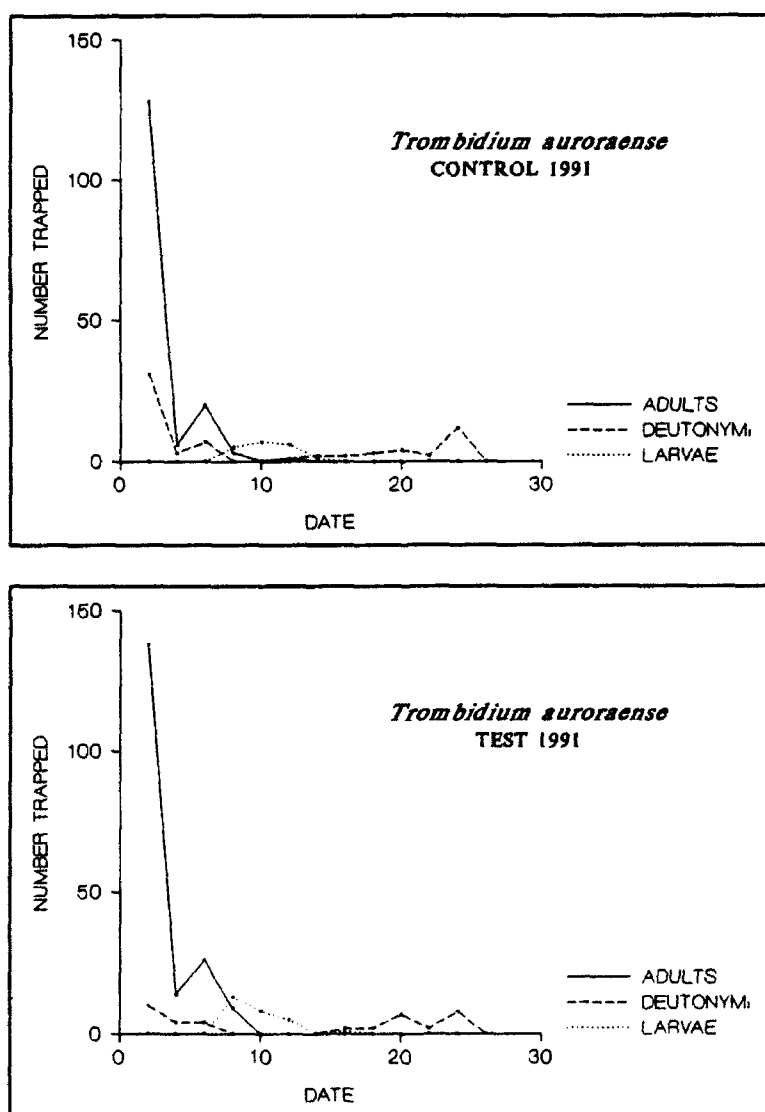


Fig. 24. Summed biweekly catches of *Trombidium auroraense*, 1991.

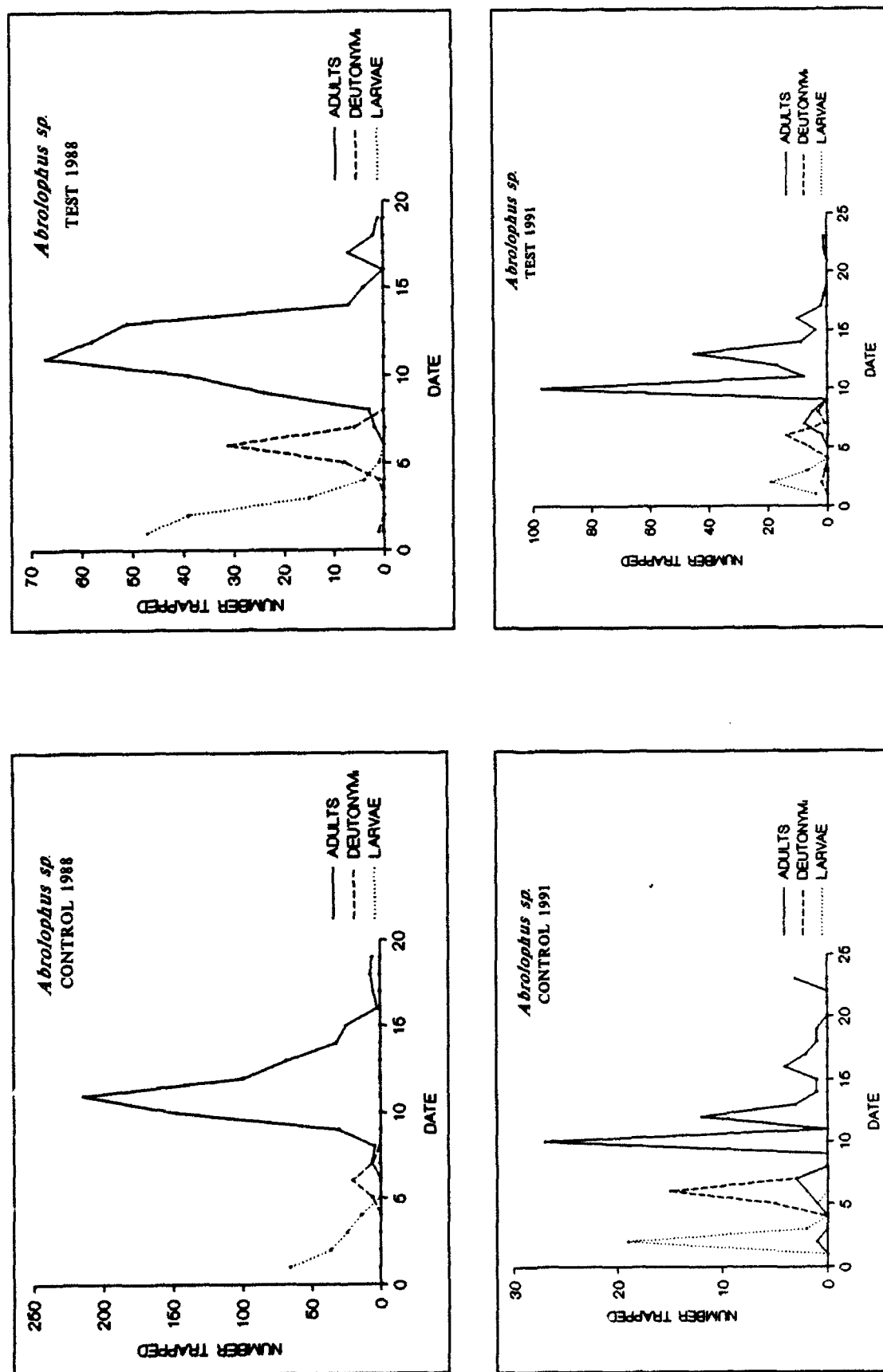


Fig. 25. Weekly catches of *Abrolophus* sp., May through September 1988 and 1991.

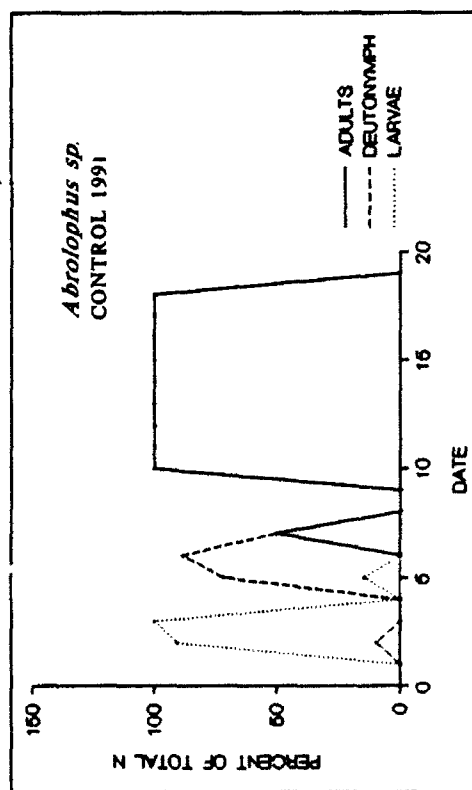
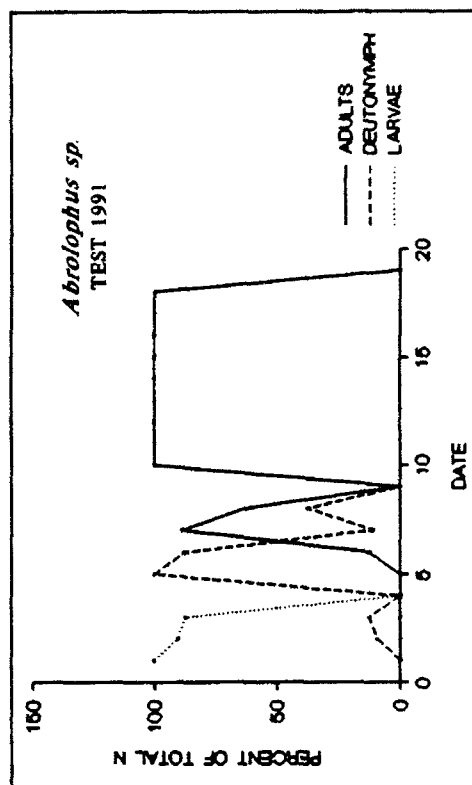
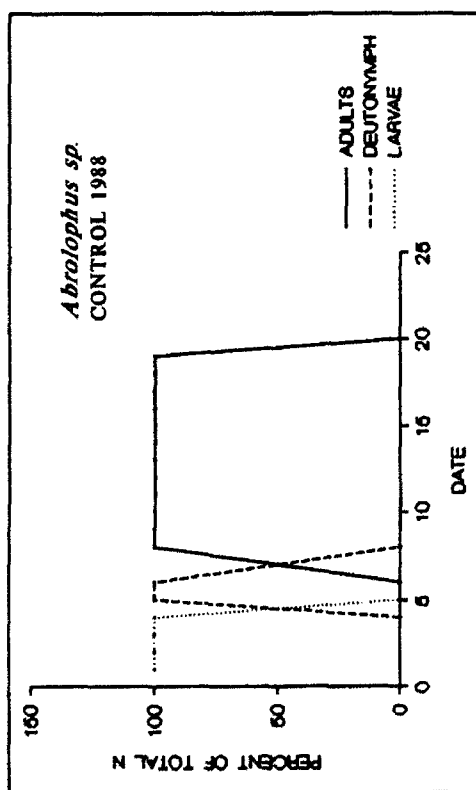
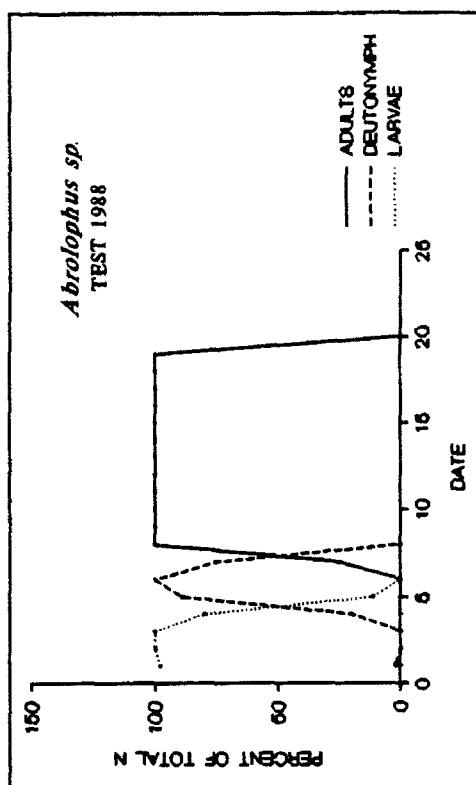


Fig. 26. Frequency of active stages (% of total N) of *Abrolophus sp.*, 1988 and 1991.

3. Carabidae:

Total numbers trapped of the most common species are listed in Table 8, providing an update through 1991. Overall totals began to decrease in 1988 (Control) or 1989 (Test) and have remained relatively low when compared to the first 3 or 4 years of the study. Much as in Collembola, year-to-year numerical variations at the single species level were observed frequently (Table 8).

Community indices for Test Carabidae did not change during operational years, but diversity of the **Control** community decreased significantly during 1989-91 (Table 9). Results of BACI analyses of [Control - Test] differences reflected this decrease, which obviously cannot be interpreted as an effect of antenna activation.

Seasonal activity patterns of dominant species during operational years were consistent with earlier observations. Data for the spring-breeding Pterostichus pensylvanicus show that an unusually high frequency of teneral adults in October 1991 occurred in both sites (Fig. 27). Numbers trapped of three summer-breeding species in 1991 (Fig. 27) illustrate our overall conclusion that ELF activation did not affect patterns of activity and development.

Table 9. Total annual catches of the most common species of Carabidae in Test and Control, 1985-1991.

	T E S T							C O N T R O L						
	1985	1986	1987	1988	1989	1990	1991	1985	1986	1987	1988	1989	1990	1991
<u>P. melanarius</u>	1087	1163	643	1528	1066	851	504	183	222	356	299	161	177	118
<u>P. coracinus</u>	146	163	134	61	85	70	120	263	450	335	306	321	303	262
<u>P. pennsylvanicus</u>	206	179	102	74	93	135	77	278	247	176	130	122	105	122
<u>P. adstrictus</u>	19	6	11	3	2	0	0	253	172	106	18	0	0	0
<u>P. mutus</u>	232	203	210	102	153	182	300	24	15	26	12	57	25	58
<u>Calathus</u> spp.	81	40	32	23	31	41	76	290	157	130	71	72	44	34
<u>C. frigidum</u>	67	139	406	132	12	2	2	29	107	185	31	5	6	4
<u>S. impunctatus</u>	103	261	104	74	49	145	205	700	894	367	157	88	293	471
<u>H. fuliginosus</u>	76	139	71	124	79	88	80	55	116	61	88	83	84	101
TOTAL ALL SPP.	2168	2506	1913	2261	1744	1637	1499	2307	2639	1936	1222	996	1162	1345
TOTAL N SPECIES	21	20	20	24	24	23	24	20	20	18	20	18	20	19

Note: Calathus spp. consist of C. ingratus and C. gregarius, and may in fact represent a single, variable species.

Table 10. Diversity and equitability indices for Test and Control carabid communities, based on diurnal, nocturnal and total catches in 1985-88 and 1989-91 (means \pm SD, N dates pre-ELF = 95, N dates operational = 73; P values based on t-tests of means within each site, and on mean differences between sites [BACI]; only dates when >1 species in both sites were present are included).

	TEST			CONTROL			BACI
	1985-88	1989-91	P=	1985-88	1989-91	P=	P=
Diurnal							
H'	1.11 \pm 0.51	1.08 \pm 0.51	NS	1.21 \pm 0.40	1.03 \pm 0.31	0.004	0.04
S'/S	0.43 \pm 0.13	0.43 \pm 0.13	NS	0.50 \pm 0.11	0.53 \pm 0.11	NS	NS
Nocturnal							
H'	1.18 \pm 0.43	1.17 \pm 0.34	NS	1.42 \pm 0.38	1.31 \pm 0.39	NS	NS
S'/S	0.40 \pm 0.12	0.43 \pm 0.13	NS	0.44 \pm 0.10	0.46 \pm 0.89	NS	NS
Total							
H'	1.32 \pm 0.51	1.27 \pm 0.44	NS	1.55 \pm 0.40	1.36 \pm 0.42	0.003	0.04
S'/S	0.38 \pm 0.17	0.41 \pm 0.16	NS	0.43 \pm 0.14	0.44 \pm 0.13	NS	NS

Variations in sex ratios and diel activity preferences have not yet been analysed in detail. However, yearly summaries show that the degree of variability encountered during operational periods generally falls within the range observed in 1985-88.

With respect to diel preferences, a few species proved more flexible than most (Table 10: *P. melanarius*, *P. pensylvanicus*, *P. coracinus*). *Harpalus fuliginosus* and *Clivina fossor* were mainly day-active in all years, *Notiophilus aeneus* was very strictly diurnal, and *Cymindis cribricollis* strictly nocturnal. One unusual occurrence (in both sites) concerns *Calosoma frigidum*: diurnal activity declined sharply in 1988 (Table 10), concurrent with a complete absence of developed eggs in females. Since then, the species has virtually disappeared (Table 8).

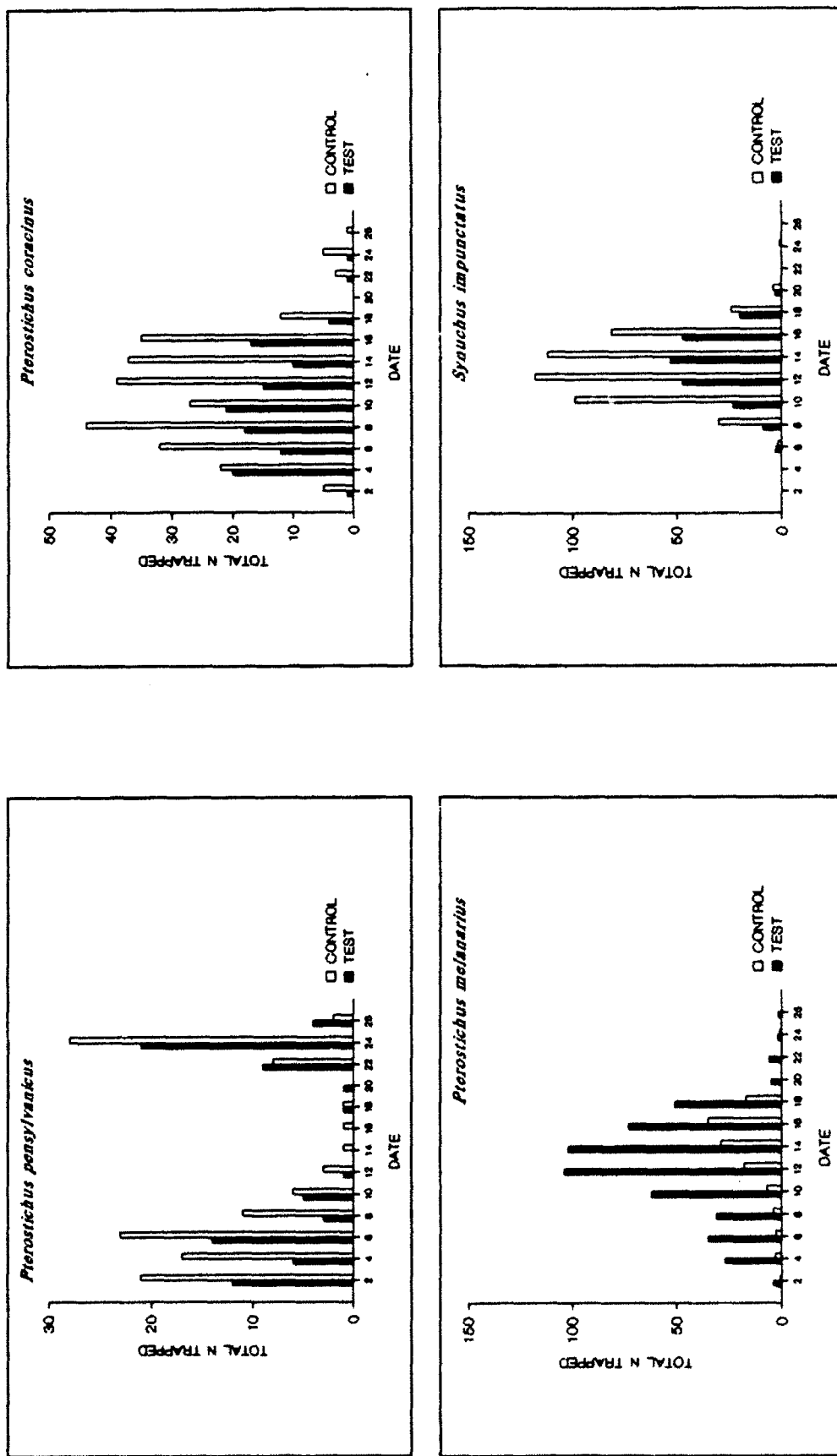


Fig. 27. Weekly catches of four carabid species in Test and Control, May through October 1991 (day and night catches summed).

Table 11. Diel activity preferences of common species of Carabidae in Test and Control, expressed in percent diurnality [(day catch / total catch per year) x 100].

	T E S T							C O N T R O L						
	1985	1986	1987	1988	1989	1990	1991	1985	1986	1987	1988	1989	1990	1991
<u>P. melanarius</u>	71.8	47.7	34.7	38.6	49.6	71.3	30.7	73.8	50.0	31.2	39.1	27.3	58.2	39.8
<u>P. pensylvanicus</u>	33.5	26.8	31.4	16.2	30.1	63.0	44.2	36.0	33.6	32.4	18.5	33.6	34.3	48.4
<u>P. coracinus</u>	55.5	27.6	22.4	14.5	29.4	40.0	34.2	60.5	38.4	25.3	5.9	18.1	28.7	23.7
<u>S. impunctatus</u>	50.5	37.9	22.1	36.5	36.7	31.7	27.3	36.4	31.9	34.1	33.1	69.3	35.8	31.8
<u>H. fuliginosus</u>	86.8	82.7	81.7	82.3	78.5	87.5	70.0	89.1	87.1	85.2	82.9	68.7	69.0	74.2
<u>N. aeneus</u>	100	100	100	97.6	100	100	90.6	95.7	100	97.2	100	100	100	-
<u>C. cribricollis</u>	0	0	0	0	6.4	9.5	9.5	0	0	1.5	2.2	2.9	0	0
<u>C. frigidum</u>	98.5	85.6	80.8	56.1	-	-	-	86.2	86.0	71.4	38.7	-	-	-
<u>C. fossor (Test)</u>	89.8	71.4	80.8	83.3	86.4	75.0	78.3	-	-	-	-	-	-	-

Year-to-year variations in sex ratios (Table 11) also showed no definite patterns which could be related to antenna activation. Overall, males were captured more frequently than females of most species, although sex ratios occasionally (to less than 50% males) in either site. Of the five species listed in Table 11, four shared the following pattern (in both sites): the lowest proportion of males occurred in 1988, a year distinguished by the longest, most severe drought among the 7 years of study. We have yet to explore potential links between weather patterns, sex-specific activity preferences, and observed sex ratios.

Table 12. Percent males (of the yearly total) among commonly trapped Carabidae, 1985-1991

	T E S T							C O N T R O L						
	1985	1986	1987	1988	1989	1990	1991	1985	1986	1987	1988	1989	1990	1991
<u>P. melanarius</u>	61.6	59.6	68.1	57.8	70.8	71.4	82.5	73.1	70.9	87.5	66.8	72.7	78.5	80.5
<u>P. pensylvanicus</u>	61.3	59.2	51.9	38.9	41.9	59.3	54.5	53.8	70.4	70.7	44.5	61.5	62.9	55.7
<u>P. coracinus</u>	58.9	64.8	62.9	55.4	67.1	75.7	76.7	68.3	65.2	61.9	51.2	60.0	73.3	63.7
<u>S. impunctatus</u>	83.2	68.6	72.9	53.3	67.3	70.3	66.8	46.4	49.9	50.0	44.7	54.5	50.2	51.6
<u>H. fuliginosus</u>	48.7	49.6	49.3	57.3	57.0	64.8	52.5	69.1	63.8	55.4	53.5	48.2	50.0	51.5

V. EARTHWORMS

1. Density and biomass

This section provides an updated summary of population trends through 1992.

In the Test site, density of Dendrobaena octaedra reached an all-time low in 1992 (Fig. 28). The species is now so scarce that data analyses have become meaningless. It is possible that this decline is related to increased numbers of Lumbricus rubellus rather than to climatic factors, since none of the last three years were limiting in terms of moisture conditions.

Lumbricus rubellus has maintained stable numbers in 1991 and 1992, following an unusually high influx of recruits in 1990. Mean biomass of the species increased significantly in 1992 (Fig. 28), due to growth of those 1990 recruits to subadult and adult size.

Aporrectodea tuberculata numbers as well as biomass declined in 1992, while A. longa remained true to its long-term history of numerical stability (Fig. 28).

In Control, populations of D. octaedra and A. turgida have been relatively high and stable for the past three years (Fig. 29), both species apparently responding to favorable edaphic conditions with good survival and high reproductive rates.

In the following, we restrict presentation of results to those of immediate concern to project goals.

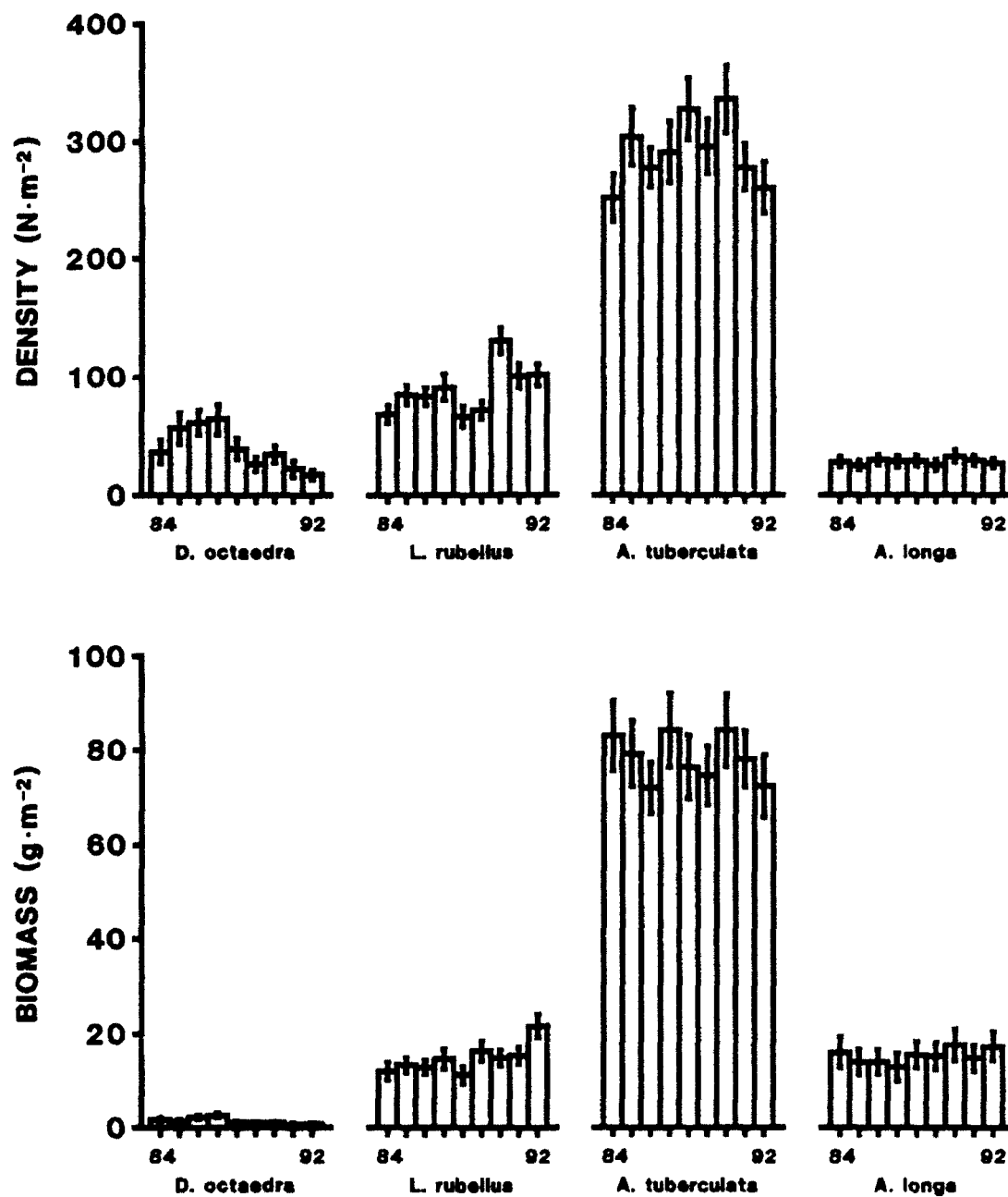


Fig. 28. Mean annual density and biomass (\pm 95% CL) of the four most common species of the Test site lumbricid community, 1984-92.

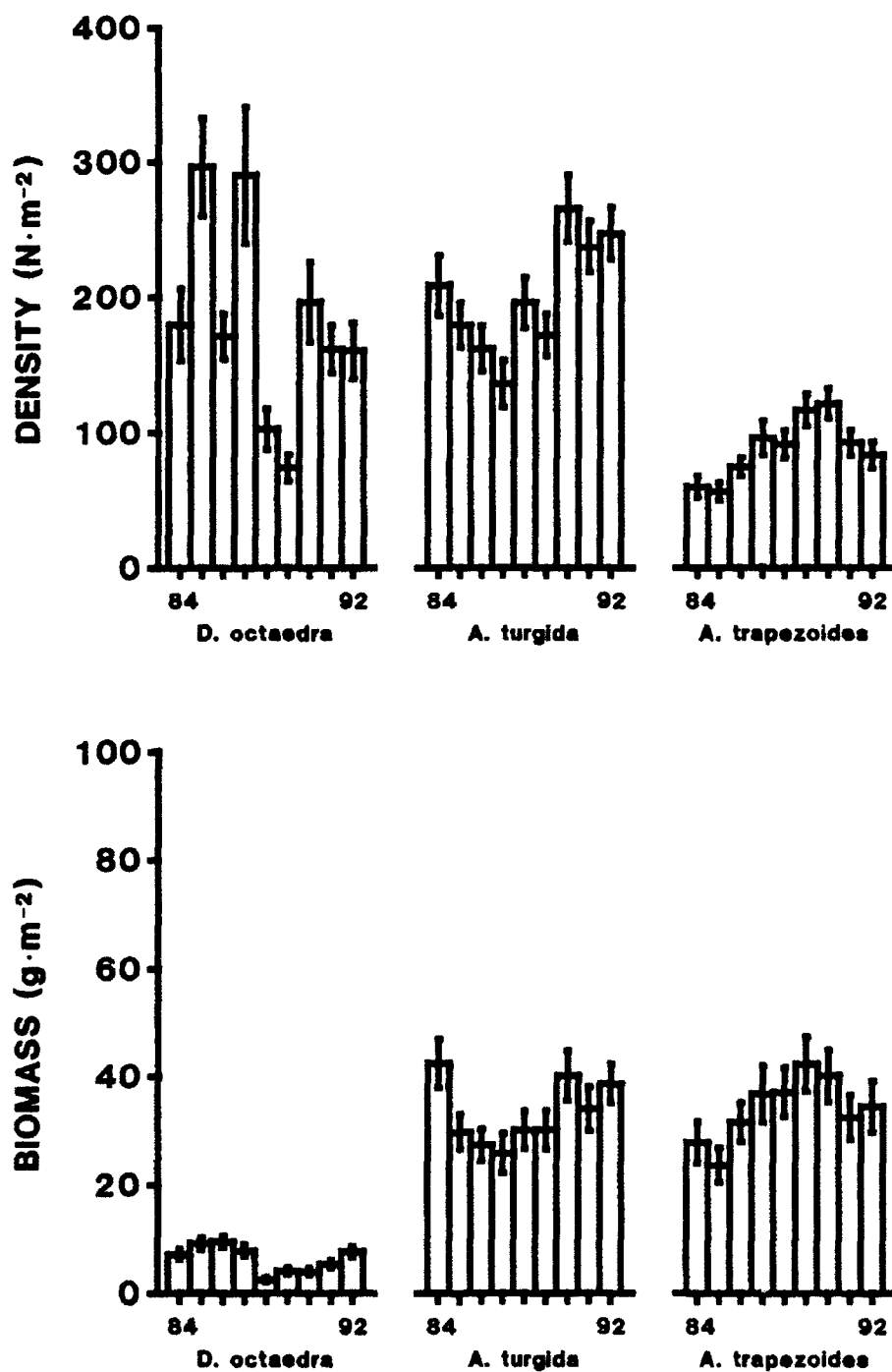


Fig. 29. Mean annual density and biomass (\pm SD) of the most common lumbricids in the Control site, 1984-92.

2. Aporrectodea tuberculata vs. A. turgida

Pre-operational data for reproductive parameters in these two species were tightly correlated (e.g., $R^2 = 0.96$ and 0.90 for clitellate proportions and cocoon densities respectively). BACI analyses of date-specific [turgida - tuberculata] differences through 1992 again indicated that reproductive performance of A. tuberculata was curtailed following ELF activation, with respect to clitellate densities, cocoon densities, and proportion of adults in the reproductive state (Table 13).

Table 13. Results of BACI tests of seasonal new cocoon densities, proportion of adults clitellate, and clitellate densities (Control A. turgida - Test A. tuberculata).

Variable	Period	N	Mean diff.	SD	t	DF	P
Clitellate density	84-88	57	2.6702	17.8695			
	89-92	48	15.5333	16.1557	-3.838	103	0.0002
Cocoon density	84-88	57	3.3053	17.1904			
	89-92	48	22.0250	21.6027	-4.944	103	0.0000
Prop. clitellate	84-88	57	0.1824	0.2053			
	89-92	48	0.2823	0.1513	-2.791	103	0.006

A series of simple illustrations aid interpretation of these results. Mean annual abundance of total adults has fluctuated somewhat in both species, that of clitellates has varied in response to climatic conditions (Fig. 30). Clitellate A. turgida showed consistently high numbers in 1989-92, but A. tuberculata did not exhibit a similar response to propitious moisture

conditions (ref. section II.2., Table 3). The same trend was evident in annual abundances of new cocoons (Fig. 31).

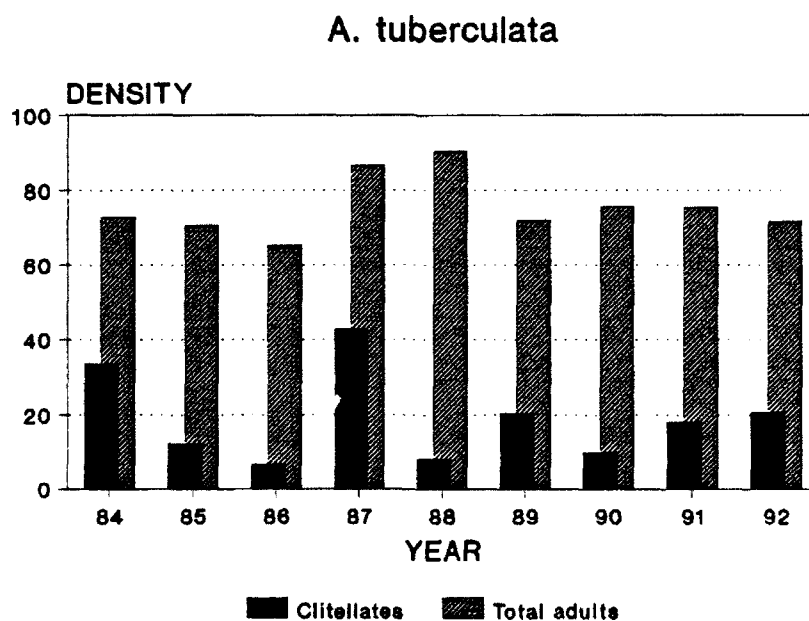
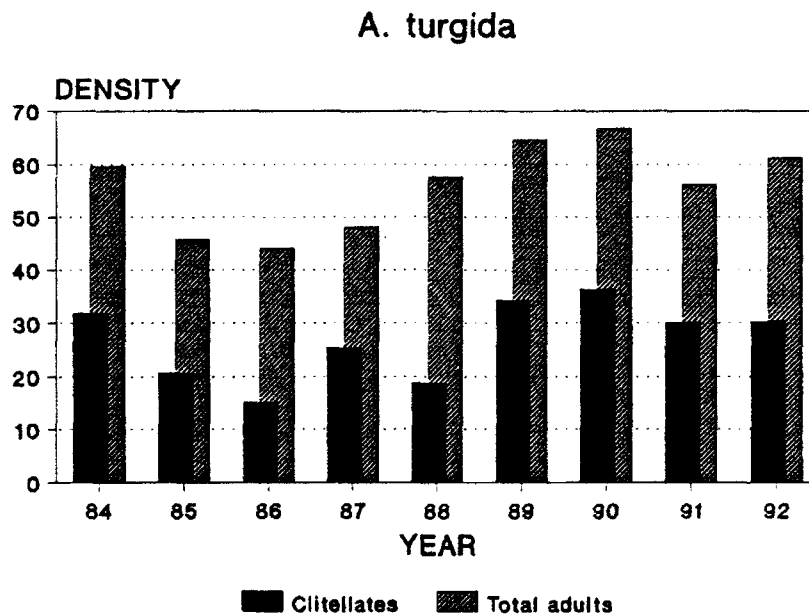


Fig. 30. Mean annual density of clitellate and total adults of *A. turgida* and *A. tuberculata*, 1984-1992.

A. tuberculata and A. turgida
New cocoons

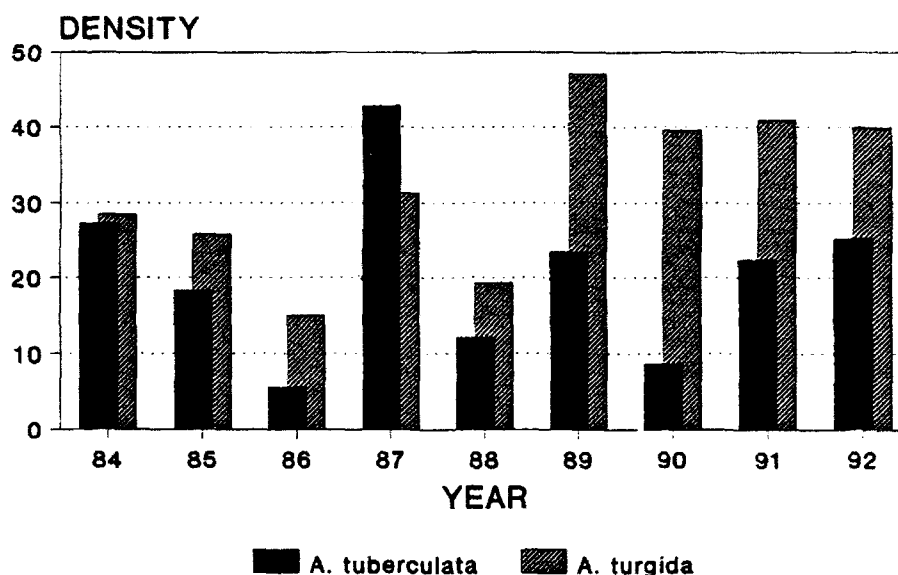


Fig. 31. Mean annual density of new cocoons of A. tuberculata and A. turgida, 1984-1992.

Annual mean differences [turgida - tuberculata] in reproductive parameters reflect these changed relationships between the species (Fig. 32). Where differences were close to zero or negative, A. tuberculata outdid A. turgida (e.g., in 1987). Large positive differences were most apparent after 1989. In all of these illustrations (Figs. 30-32) it is clear that the most "aberrant" data were obtained in 1990, the year following antenna operation. Nature or magnitude of the response to EM fields thus appears to change over time, initial effects being most pronounced.

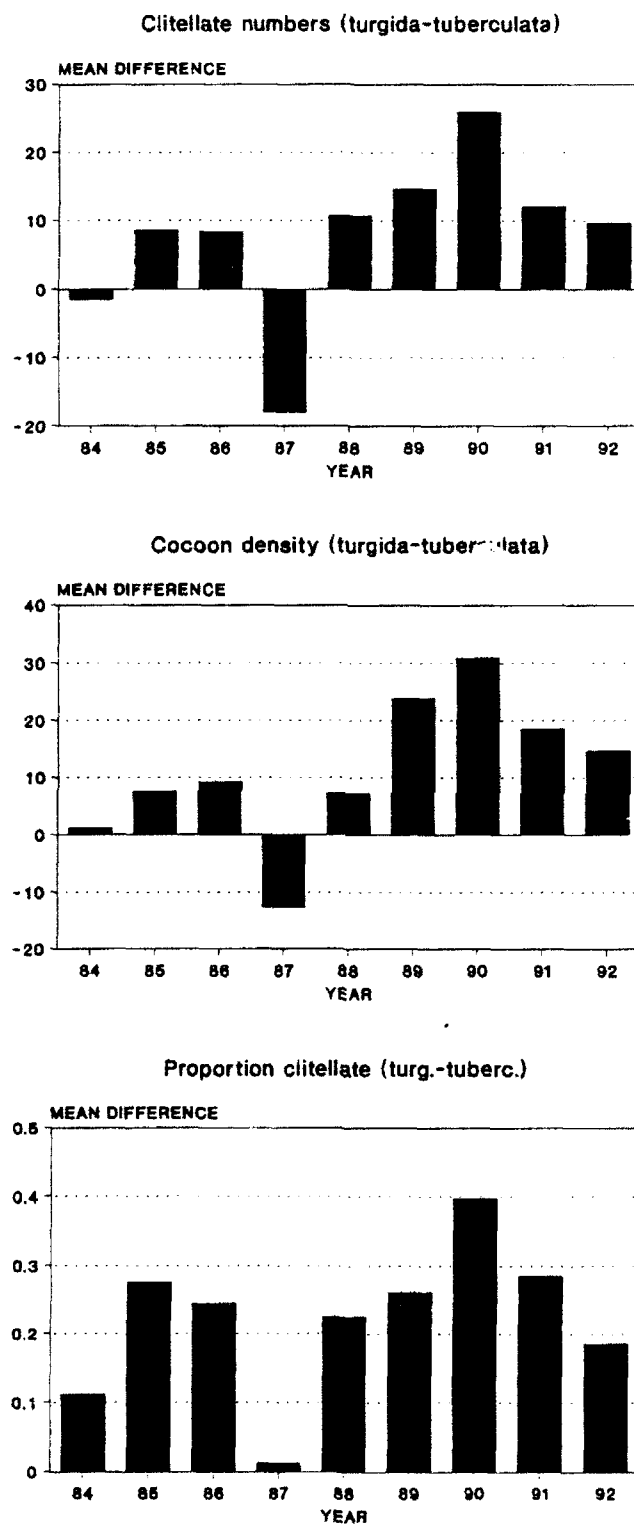


Fig. 32. Mean annual differences [turgida - tuberculata] in reproductive parameters, 1984-1992.

Finally, prompted by results of earthworm isolation experiments (section V.4.), we examined cocoon/clitellate ratios in the two species. Two methods of calculation were used:

Method 1:

$$\text{Ratio} = \{N \text{ cocoons}_{\text{date } i} / [(N \text{ clitell.}_{\text{date } i} + N \text{ clitell.}_{\text{date } i-1}) / 2]\},$$

which relates an "average" number of clitellates per 2-week period to the number of cocoons found at the end of that period.

Method 2:

$$\text{Ratio} = N \text{ cocoons}_{\text{date } i} / N \text{ clitellates}_{\text{date } i},$$

which simply relates cocoon and clitellate abundance on each sampling date.

Cases where the total (or average) number of clitellates was <3 were excluded. Date 1 of each year was also excluded, since cocoons which look "new" in early May may have been produced the preceding fall.

Mean differences [turgida - tuberculata] in cocoon/clitellate ratios decreased drastically during 1989-92, by factors of approximately 4 to 5 (Fig. 33); i.e., cocoon production rates by A. tuberculata **increased** following ELF activation. When the data were broken down into "seasonal" groups encompassing approximately 6 weeks, appreciable differences between pre-ELF and operational periods again became apparent (Fig. 34). However, none of the means, seasonally or yearly, were significantly different. Field-derived data harbor relatively high variances, and occasionally low replication (in the case of seasonal estimates, particularly during dry periods).

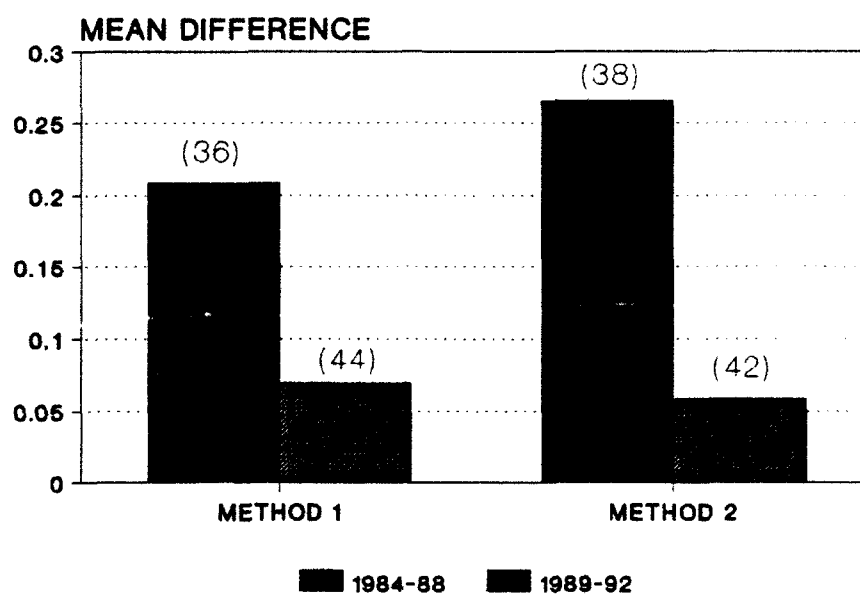
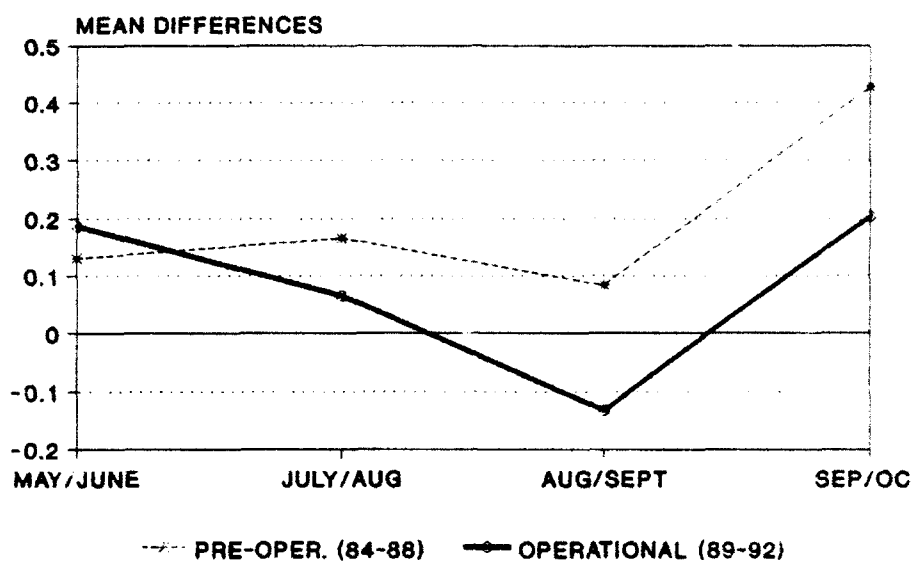


Fig. 33. Mean differences [turgida - tuberculata] in cocoon/clitellate ratios during pre-ELF and operational periods; N observations in parentheses; for explanation of Methods, refer to text.

METHOD 1



METHOD 2

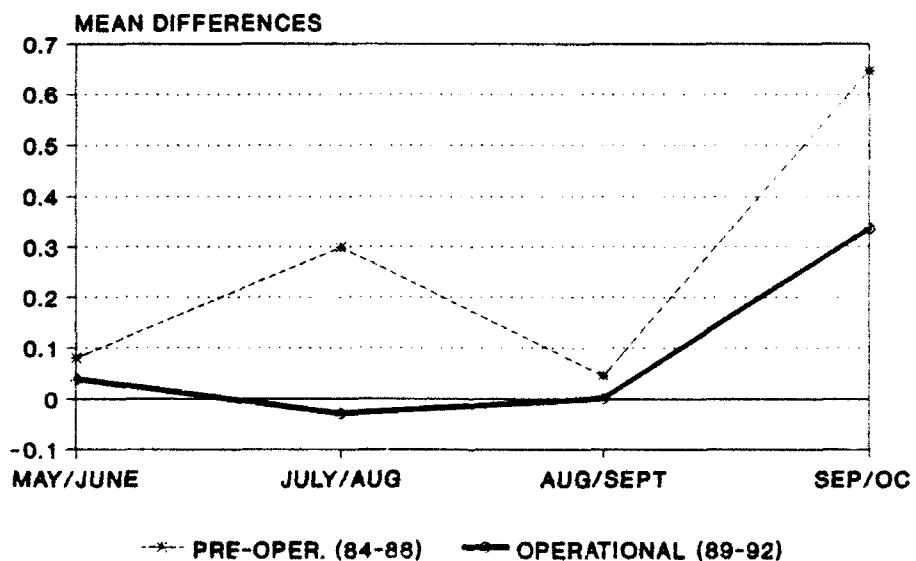


Fig. 34. Mean differences [*turgida* - *tuberculata*] in seasonal cocoon/clitellate ratios during pre-ELF and operational periods. For explanation of Methods, refer to text.

3. Aporrectodea tuberculata

A. Vertical distribution

Multiple regression of the proportion of the population in the A horizon on edaphic variables showed that A horizon moisture and temperature were influential factors, but not B horizon moisture. As discussed previously, the explanatory power of these analyses is relatively poor, although all are significant (Table 14). The lowest R^2 (0.30) was obtained for the 1989-92 period, but this is not believed to be a reflection of potential ELF effects. Time periods during which vertical migration occurred frequently tend to yield higher coefficients (1984-86, $R^2 = 0.64$). Those in which soil moisture remained above a stress-threshold (approximately 18 to 20 percent, e.g., 1989-92) did not yield high coefficients of determination. Indeed, low R^2 can be taken as indicative of low incidence of moisture stress.

Table 14. Anova table for regression analysis of proportion of A. tuberculata in the A horizon on A horizon moisture and temperature.

Source	Period	SS	DF	MS	F-ratio	P	R^2
Regression	84-86	0.6905	2	0.3452	29.31	0.000	0.64
Residual		0.3887	33	0.0118			
Regression	87-88	0.4720	2	0.2360	7.68	0.004	0.46
Residual		0.5532	18	0.0307			
Regression	89-92	0.2892	2	0.1446	9.56	0.000	0.30
Residual		0.6802	45	0.0151			

B. Clitellate densities and proportion clitellate

The size of the total adult pool has remained relatively high over the years (Fig. 30), so that the number of individuals available for entry into the reproductive sub-pool should not be a factor determining clitellate numbers.

For clitellate abundance as well as for proportion (of all adults) in the reproductive state, multiple regression again showed significant results for all three periods, but much reduced R^2 for operational years (Tables 15-16).

It may be noteworthy that R^2 based on 1989-1991 data was 0.14 and 0.27 for clitellate densities and proportions respectively (Annual Report for 1991). When 1992 data were included, R^2 rose slightly, to 0.26 and 0.34 respectively (Tables 15-16). In the case of clitellate densities, the relationship with soil moisture had lost its significance ($P = 0.45$ in 1989-1991), but P rose to 0.07 after inclusion of 1992 in the operational data subset, indicating a partial return to pre-ELF reproductive behavior.

Table 15. Anova table for regression of A. tuberculata clitellate abundance on lagged abundance and A horizon moisture.

Source	Period	SS	DF	MS	F-ratio	P	R^2
Regression	84-86	7508.7	2	3754.4	80.92	0.000	0.84
Residual		1391.8	30	46.4			
Regression	87-88	6809.6	2	3404.8	21.32	0.000	0.73
Residual		2555.1	16	146.8			
Regression	89-92	1291.1	2	645.5	7.06	0.002	0.26
Residual		3750.0	41	91.5			

Table 16. Anova table for regression of clitellate proportions of A. tuberculata on lagged proportions and A horizon moisture.

Source	Period	SS	DF	MS	F-ratio	P	R ²
Regression	84-86	1.3396	2	0.6698	59.09	0.000	0.80
Residual		0.3401	30	0.0113			
Regression	87-88	0.7935	2	0.3967	45.66	0.000	0.85
Residual		0.1390	16	0.0087			
Regression	89-92	0.2848	2	0.1424	10.55	0.002	0.34
Residual		0.5534	41	0.0135			

Mean monthly clitellate abundance (Fig. 35) illustrates the reasons for our tentative conclusion. Numbers of reproductive adults were uniformly depressed during the second half of 1989 and throughout 1990. Beginning in 1991, this "flat profile" of reproductive activity gave way to a modulated one reminiscent of moisture-dependent seasonal variations observed in pre-ELF years (Fig. 35). Given the relatively high moisture conditions in 1989-1992, we still maintain, however, that reproduction (as measured by numbers of clitellate adults) has so far remained below the species' potential (ref. 1984 and 1987 in Fig. 35; and section V.2., Table 3).

C. Cocoon production

Seasonal patterns of cocoon deposition (Fig. 36) showed clear similarities with those of clitellate abundance (Fig. 35), changing from a "flat" to a modulated profile after the second year of antenna operation.

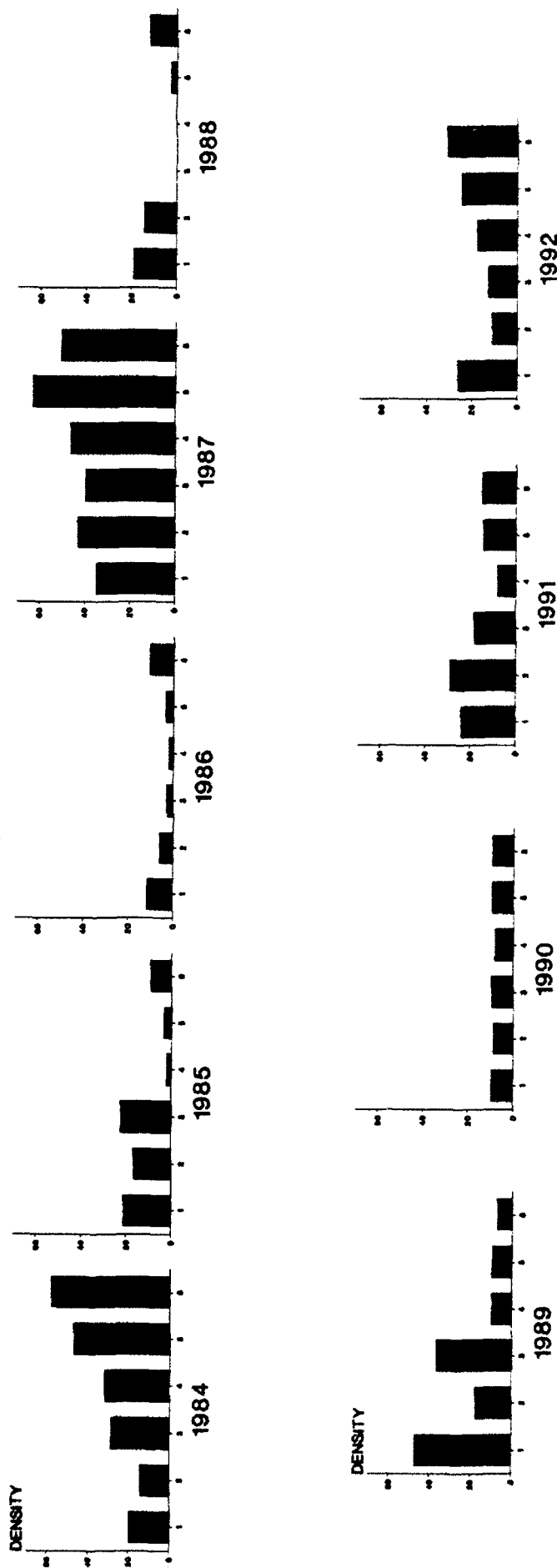


Fig. 35. Average monthly densities of clitellate adult *A. tuberculata*, 1984-1992. (Month 1 = May, Month 6 = September/October).

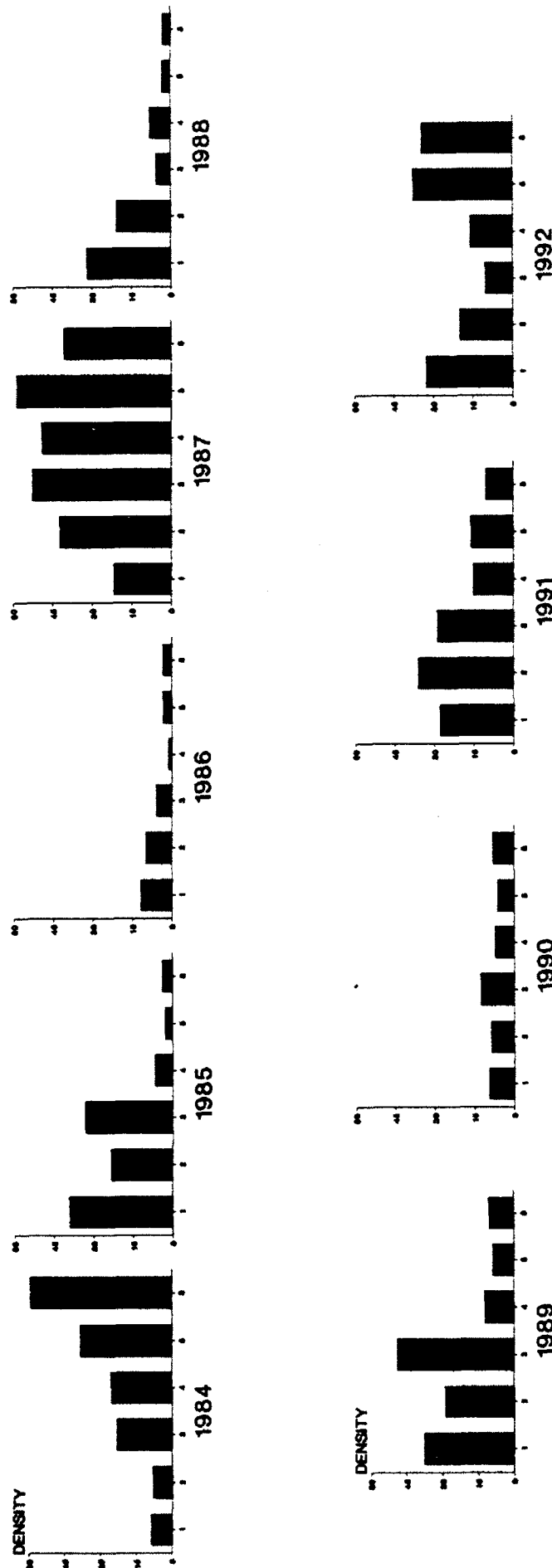


Fig. 36. Average monthly densities of *A. tuberculata* new cocoons, 1984-1992. (Month 1 = May, Month 6 = September/October).

Multiple regressions of cocoon on clitellate densities were highly significant, although R^2 for operational years was much lower than for preceding years (Table 17). In order to investigate the reasons for this decline, we calculated cocoon production rates by the two methods detailed in section V.2. The first sampling date of each year was again excluded, as were dates when <3 clitellates were recovered from samples.

Table 17. Anova table for regression analysis of new cocoons of A. tuberculata on clitellate density and lagged clitellate density.

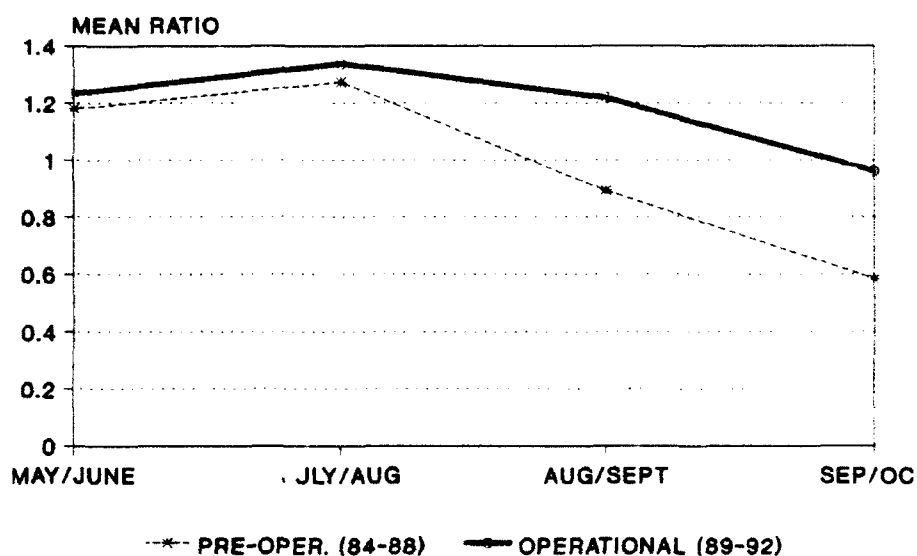
Source	Period	SS	DF	MS	F-ratio	P	R^2
Regression	84-86	7487.6	2	3743.8	76.57	0.000	0.84
Residual		1466.8	30	48.9			
Regression	87-88	8521.6	2	4260.8	56.56	0.000	0.86
Residual		1431.3	19	75.3			
Regression	89-92	3980.0	2	1990.0	23.45	0.000	0.53
Residual		3479.6	41	84.9			

Yearly mean cocoon production rates were indeed higher during operational years, but only Method 2 calculations produced significant results ($P < 0.05$) (Table 18). Summarized by 5 to 6 week seasonal periods (Fig. 37), 1989-92 ratios were consistently higher than 1984-88 ratios; both methods, however, yielded significant differences only for the September-October period ($P < 0.05$).

Table 18. Mean (\pm SD) cocoon/clitellate ratios for A. tuberculata during pre-ELF and operational periods. For explanation of Method 1 and 2 refer to text, section V.2. (N dates in parentheses).

Period	Method 1	Method 2
1984-88	1.0392 ± 0.5084 (39)	1.0156 ± 0.5645 (41)
1989-92	1.2080 ± 0.5699 (44)	1.2938 ± 0.6499 (42)

METHOD 1



METHOD 2

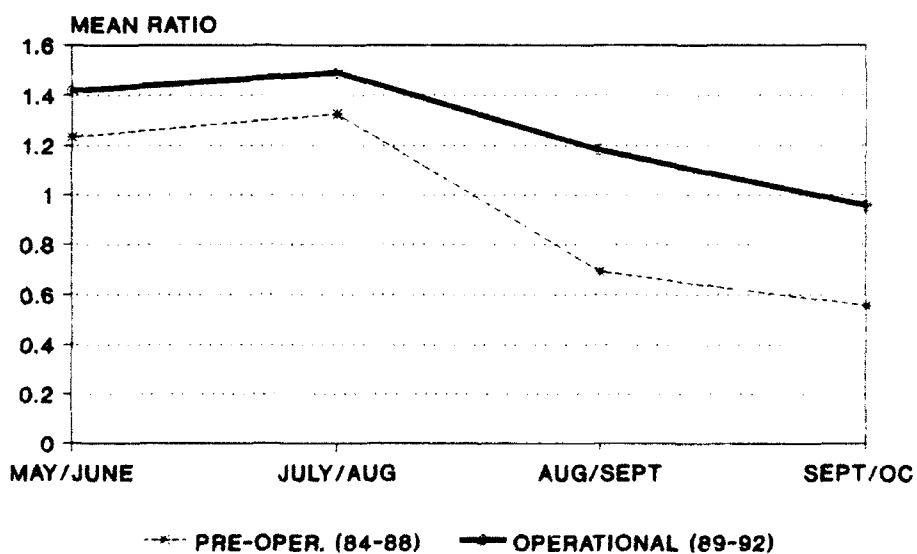


Fig. 37. Mean seasonal cocoon/clitellate ratios for *A. tuberculata* during pre-ELF and operational years. For explanation of Methods refer to section V.2.

D. Population structure

In long-lived lumbricids, consequences of reproductive events on population structure may become evident only after a delay of one or more years. We have yet to subject population structure data to rigorous analysis. Preliminary summaries show, however, that the parameter may indeed be of interest once all operational data (through 1993) are available. At this time, we present an overview for A. tuberculata as well as A. turgida, and discuss possible causes for observed variation.

During 1984-88, significant fluctuations in mean annual [immature/adult] ratios occurred in an essentially synchronous mannner in both species ($R^2 = 0.82$) (Fig. 38). High cocoon production in 1984 resulted in larger numbers of immatures in 1985 and 1986. Low cocoon production in 1986, a drought year, reduced immature numbers in 1987, but propitious moisture conditions in 1987 again produced an increase in immature proportions in 1988 (Fig.38).

Discrepancies between the species began in 1989. The severe drought of 1988 curtailed reproduction in A. turgida as well as A. tuberculata. For A. turgida, the consequences clearly entailed a significant decrease in immature proportions in 1989 (Fig. 38). Not so for A. tuberculata: large numbers of cocoons were produced during the first three months of 1989 (Fig. 36), many of which hatched yet the same year; high immature ratios in the fall of 1989 and into 1990 resulted in an increased mean ratio for both years (Fig. 38).

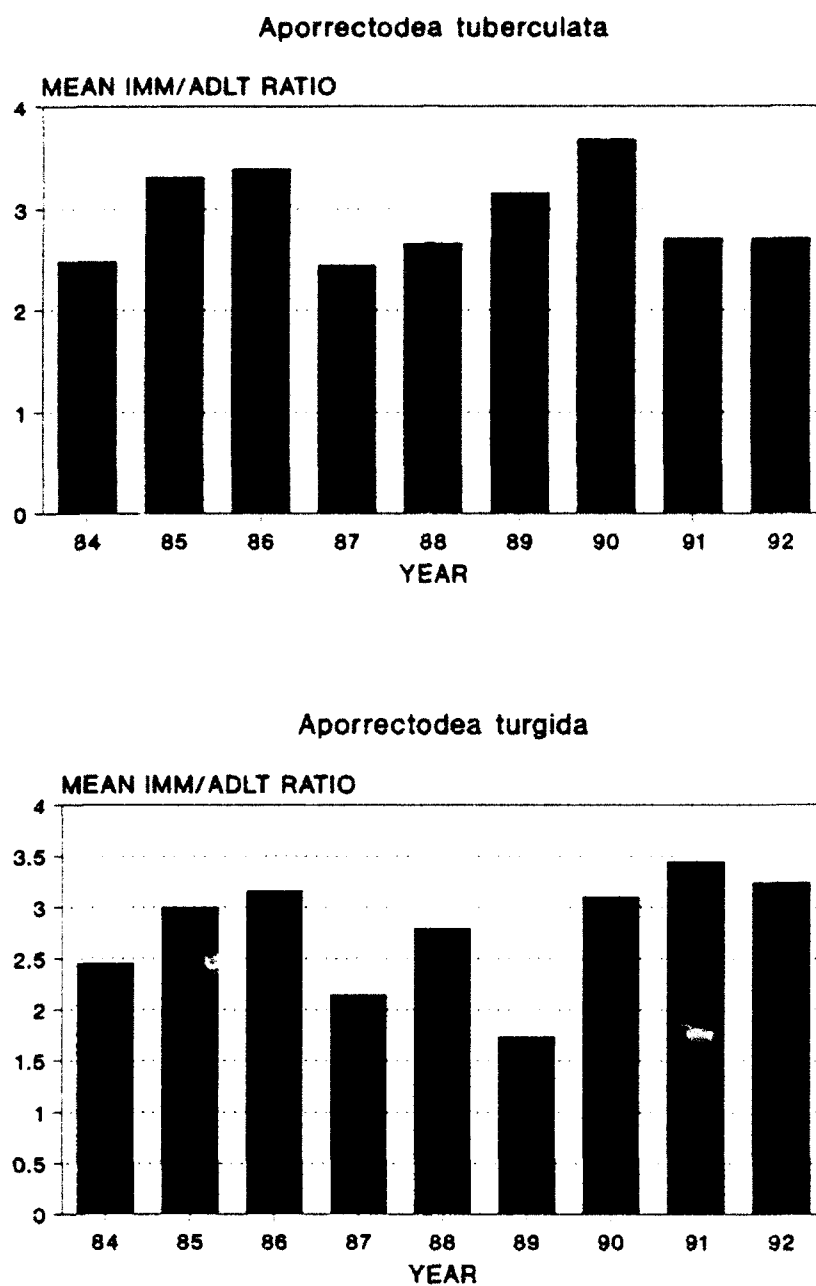


Fig. 38. Mean annual immature/adult ratios for *A. tuberculata* and *A. turgida*, 1984-1992.

We have shown that A. tuberculata reproduction was curtailed in 1990 (Figs. 35-36) despite propitious moisture conditions (Table 3, section II). Significantly lower immature proportions in 1991 were the result ($P < 0.01$), an effect which persisted into 1992 (Fig. 38). Indeed, the 1990-91 decrease was reminiscent of the 1986-87 reduction ($P < 0.001$) caused by the summer drought of 1986.

On the other hand, A. turgida responded to favorable moisture levels with high levels of cocoon production throughout the four operational years (Fig. 30), which correspondingly raised immature proportions to fairly constant high values in 1990-92 (Fig. 38).

There is thus evidence that altered reproductive patterns have detectable consequences on population structure and, in the long term, on population density. A detailed data base exists with respect to seasonal population structure (in terms of numbers and mass of individuals), but analytical methods have yet to be developed. If possible, they should take into account not only environmental variables, but estimates of developmental time and longevity as well.

4. *Aporrectodea tuberculata* isolation experiments

A. Methods and experimental design

Cylindrical fiberglass mesh bags (20 cm diameter) are buried to a depth of 20 cm in tightly fitting holes in Test and Control sites. They are filled with air-dried, sieved and remoistened soil (approx. 7 l soil dry volume per bag), which is manually compressed and settled by adding water. After a 24 hour equilibration period, A. tuberculata are added to the soil surface and covered with moist leaf litter. Bags are retrieved at monthly intervals, and earthworms and cocoons are recovered.

In 1991, bags were left undisturbed between sampling occasions. In 1992, the protocol was expanded to include:

a) Periodic soil temperature records by means of YSI telethermometer, taken approx. 1 hour apart in Test and Control bags at 5 and/or 10 cm depth. On average, temperatures in Test bags were slightly higher than in Control (range among five dates: 0.2 to 0.5°C), but the differences were far from being significant.

b) In each site, three additional bags were installed which contained TDR (Time Domain Reflectometry) sensors. Frequent readings of soil moisture were taken in these bags, and were supplemented by occasional readings from experimental bags.

These records formed the basis for a watering regime, designed NOT to keep all bags at equal moisture levels at all times, but to prevent average moisture from dropping below a threshold of approx. 18% (determined empirically as adequate for earthworm activity).

An additional six TDR bags were installed in the Control site; from them (as well as from the three Test and Control permanent TDR bags prior to retrieval at sampling time) a total of 87 records of TDR vs. gravimetric moisture content were obtained. By regression ($R^2 = 0.84$), it was determined that a TDR reading of approx. 20% corresponded to approx. 18% moisture (gravimetric).

In practice, whenever any ONE of the three TDR bags gave a reading of approx. 20%, between 0.75 and 1.0 gal water was added to each bag in both sites. This became necessary on six occasions during the entire season. In one instance when TDR readings fell slightly below 20%, watering was omitted because the next sampling was scheduled 2 days hence.

In both sites, readings from TDR and experimental bags on five dates (N varied from 6 to 15 per date) showed that mean TDR records were highly representative of conditions in experimental bags: means differed by up to $\pm 2\%$ moisture, but mostly by $\leq 1\%$ moisture.

c) In 1992, all worms were weighed individually at every sampling, without allowing them to void their intestine. In addition, a number of earthworms kept in "reference bags" (treated exactly like experimentals) were weighed and re-weighed after keeping them on wet filter paper for 3-4 days, in hopes of deriving a correction factor for "gut-voided mass" for experimental animals. However, the effort proved futile. Omitting all detail, we concluded that:

- individual earthworms stemming from the same reference bag may either lose or gain mass, or experience no change, in an

unpredictable manner; i.e., mass loss through gut content loss, and mass gain through re-hydration on wet towelling, vary in their relative magnitude between single individuals recovered from a given bag;

- despite the potential error inherent in "gut full" mass records, they seem preferable to estimates obtained by means of a badly quantifiable and questionable correction factor.

d) Experimental design: unlike 1991 (when each group of earthworms was returned to its bag of origin at each sampling), the 1992 protocol was changed as follows:

After obtaining live mass data, each group of worms was randomly redistributed such that each bag received approx. equal numbers of clitellate and non-reproductive individuals. Each incubation period thus began with approx. equal stage structure in each bag. The procedure allowed use of parametric statistics because it alleviated the problem of auto- or serial correlation.

e) Lastly, blind scoring was used for processing wormbags, so that the investigator did not know the provenance of cocoons during counting and weighing, nor of earthworms up until their redistribution (for explanation of provenance, see section B. below).

B. Earthworm provenance and replication, 1991 vs 1992

a) 1991 series: Earthworms came from two locations: the Test site, and a second site (Merriman Rd.) removed from ELF influence. After some adjustments, the protocol was finalized in mid-July 1991. From then on, replication was 5 bags, with 30 worms in each,

per provenance (Test and Merriman Rd.) Both provenance series were incubated in soil collected near the Test site. On each sampling occasion, each group of worms was returned to its bag of origin without having been weighed.

Having remained in the field over winter, the Merriman Rd. series was discontinued in late April 1992. The Test provenance series was kept on (5 replicates / site), but earthworms were randomly redistributed over the five bags after having been weighed (see section A.d. above). Unlike 1991, individuals missing or injured were not replaced (appropriate reserves were no longer available).

b) 1992 series: A new series was initiated in April-May 1992, with 600 A. tuberculata collected from yet another site removed from ELF influence ("Fire Tower" site). They were randomly divided into groups of 30, mimicking the developmental structure of the population at the time of collection (3.7% clitellate, the remainder non-reproductive).

Replication consisted of 10 bags in Control and 10 in Test. The substrate consisted of soil obtained in the Fire Tower site prior to each sampling, treated and mixed in the same way as in 1991 (70% A horizon and 30% B horizon material, air-dried and sieved).

Injured animals (their developmental state was always recognizable) were replaced with worms from reservoir bags kept in each site for that purpose. Each group of earthworms was redistributed upon sampling as described in section A.d. above.

Both series were first sampled (Test provenance 1991) or

installed (Fire Tower 1992 provenance) on May 8, 1992, then retrieved at 4 week intervals until October 22. Sampling will again resume in late April 1993.

It is important to keep in mind that A. tuberculata in the Test provenance series had been exposed to ELF fields from May 1989 to May 1991 prior to being incubated in Test and Control; and that they have now been adults for at least 2 years (half of them were post-reproductive at the time of collection in 1991) and are currently entering their third or fourth year of adulthood.

Fire Tower provenance A. tuberculata, on the other hand, had never been exposed to appreciable ELF fields prior to incubation in 1992; they are now entering their second year of exposure in Test bags, and their second (known) year of adulthood.

C. Results

C. 1. Electromagnetic fields

In 1991, field intensities inside wormbags were measured in late May, after the bags had been in place for 10 days. Measurements were repeated in May 1992. It was established in both years that electric fields in Control were lower than those in Test by a factor of ≥ 250 .

On average, electric field intensities in 1992 were almost double those recorded in 1991 (Table 19), reduction of fields with respect to those outside of bags dropping from 53% to 28%.

We believe that increased field intensities in the second year of the study were mainly due to continued use of the same locations

(holes) in which wormbags were installed. By repeating the watering-in process on each sampling occasion, minor discontinuities in the hole walls were smoothed out and contact with surrounding soil (and thus penetration by electric fields) was improved.

Table 19. Average (\pm SD) electric field intensities in and around wormbags in Test and Control, 1991 and 1992.

	Earth electric field intensities (mV/m)	
	Within bag	Next to bag
1991 Test	24.0 \pm 6.7	54.0 \pm 7.2
Control	0.094 \pm 0.044	0.21 \pm 0.13
1992 Test	46.0 \pm 10.6	64.0 \pm 11.2
Control	0.144 \pm 0.036	0.186 \pm 0.035

C. 2. Earthworm body mass

Among A. tuberculata of Test provenance, mean body mass of clitellate individuals did not differ significantly, although non-reproductive worms (as well as all stages together) were heavier when incubated in Control ($P < 0.001$) (Table 20). ANOVA showed main effects as well as interactions to be highly significant (Table 21). Presence of an interaction points out a shift in Test/Control relationships during the second half of the season, when earthworms were of approximately equal mass in both sites (Fig. 39). For clitellates, no clear seasonal between-site differences were observed (Fig. 40).

For A. tuberculata of Fire Tower provenance, mean mass of worms

incubated in Test exceeded that of Control individuals, be it for separate or combined developmental stages (Table 20, Figs. 39-40). The data for the entire season did not lend themselves to ANOVA, but single-date analyses showed that worms incubated in Test bags were significantly heavier than Control worms on all dates, except for the May 8 initial incubation date.

We do not know whether these results are biologically meaningful, in view of the potential error inherent in "gut full" mass estimates. We will continue monitoring body mass, in the belief that the second year of exposure (Fire Tower series) may allow more definite conclusions.

Table 20. Mean body mass (\pm SD) of earthworms of Test and Fire Tower provenance during the 1992 season. (N observations in parentheses).

Provenance	Mean mass (mg) \pm SD			
	Test site		Fire Tower site	
Incubat. site	Test	Control	Test	Control
Clitellate	876.1 \pm 169.8 (190)	892.0 \pm 184.6 (334)	1118.8 \pm 239.0 (1036)	1031.0 \pm 216.3 (974)
Non-reprod.	682.2 \pm 196.3 (772)	732.4 \pm 179.6 (658)	901.1 \pm 215.5 (970)	882.0 \pm 204.9 (1090)
All worms	720.5 \pm 209.4 (962)	786.1 \pm 197.7 (992)	1013.6 \pm 256.7 (2006)	952.3 \pm 228.3 (2064)

Table 21. Analysis of variance table for body mass of A. tuberculata, Test Provenance series, during the 1992 season (May through October).

Source	SS	DF	MS	F-ratio	P
Date	0.8522E+07	5	0.1704E+07	44.64	0.0000
Site	0.1434E+07	1	0.1434E+07	37.56	0.0000
Date x site	985328.87	5	197065.77	5.16	0.0001
Error	0.6269E+08	1642	38177.11		

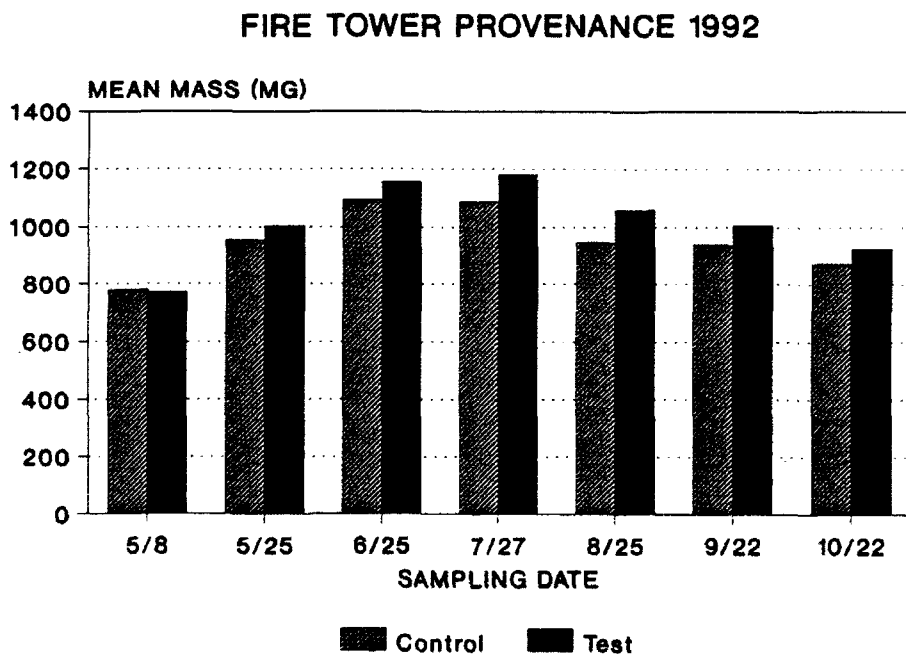
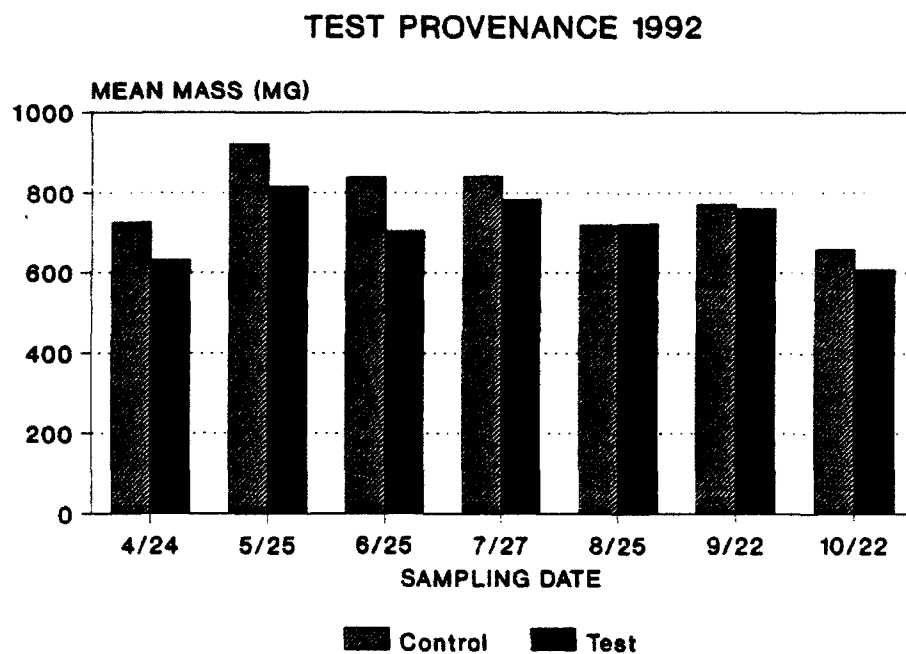


Fig. 39. Mean body mass (mg) of *A. tuberculata* of Test Provenance (initiated May 1991) and Fire Tower Provenance incubated in wormbags during 1992.

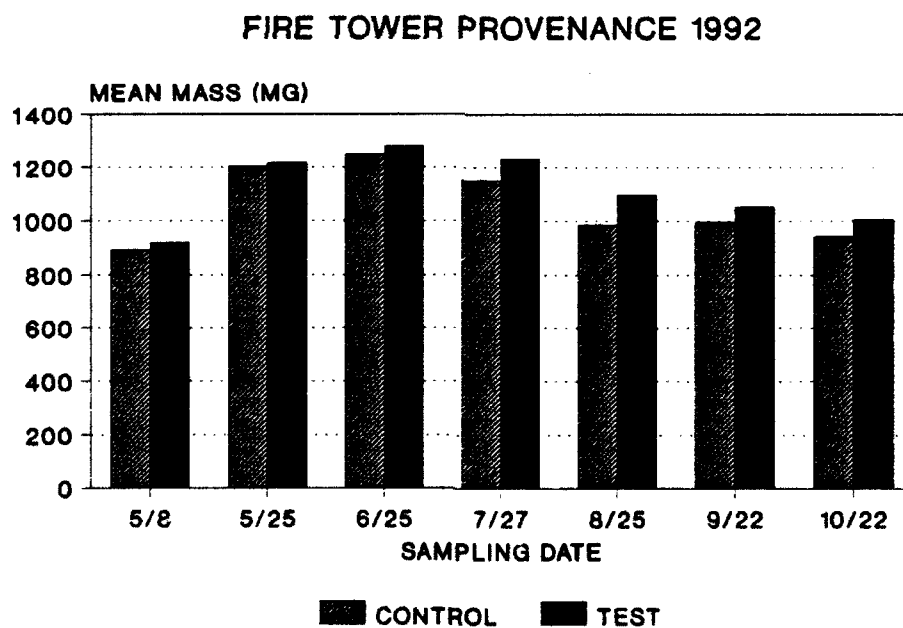
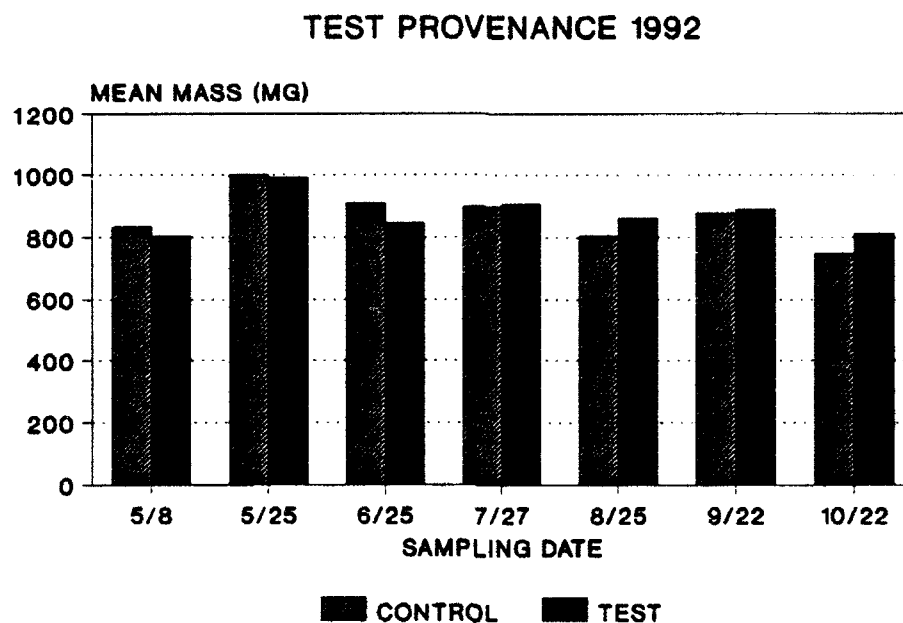


Fig. 40. Mean body mass (mg) of clitellate *A. tuberculata* of Test (series initiated May 1991) and Fire Tower Provenance, incubated in Test and Control in 1992.

C.3. Cocoon mass

Within each provenance series, mean mass of individual cocoons did not differ between incubation sites (Table 22). Within each site, however, worms of Test provenance produced significantly smaller cocoons than those of Fire Tower provenance ($P < 0.001$). It is likely that lower body mass (Table 20) and production of correspondingly smaller cocoons (Table 22) are evidence of senescence, and that Test provenance individuals are approaching the end of their life span.

Table 22. Mean (\pm SD) mass of cocoons produced by *A. tuberculata* of Test and Fire Tower site provenance, incubated in Test and Control in 1992. (N cocoons weighed in parentheses).

Provenance	Mean mass (mg) \pm SD	
	Test site	Fire Tower site
Incubation site		
Test	22.335 \pm 3.825 (143)	25.239 \pm 4.012 (884)
Control	22.973 \pm 3.826 (252)	24.879 \pm 3.896 (659)

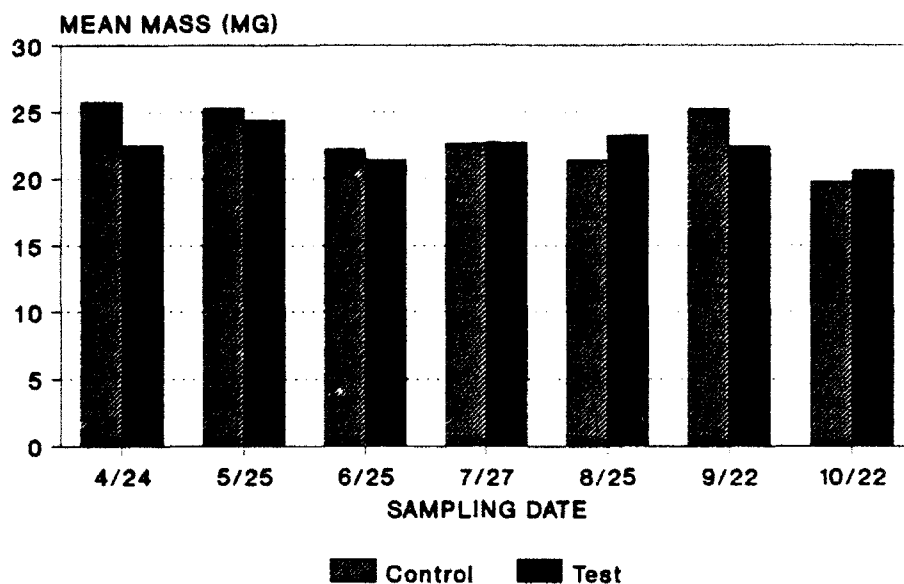
Although date effects were significant for both provenance series, no site effects were evident (Table 23). Graphically presented, the data clearly show that cocoons of similar mass were produced in the presence or absence of EM fields. Occasional

significant differences between date-specific means (e.g., Fire Tower series, June 25, and Test series, Sept. 22, Fig. 41) were always associated with low replication and are unlikely to have biological significance.

Table 23. Results of ANOVA of average mass of cocoons deposited during 1992 by A. tuberculata incubated in Test and Control (Test and Fire Tower provenance).

Source	SS	DF	MS	F-ratio	P
Test series					
Date	276.95	5	55.39	4.18	0.0011
Site	3.30	1	3.30	0.25	0.6182
Date x site	118.63	5	23.73	1.79	0.1139
Error	4609.82	348	13.25		
Fire Tower series					
Date	1567.23	4	391.81	26.87	0.0000
Site	5.49	1	5.49	0.38	0.5395
Date x site	91.08	4	22.77	1.56	0.1821
Error	22339.36	1532	14.58		

TEST PROVENANCE 1992



FIRE TOWER PROVENANCE 1992

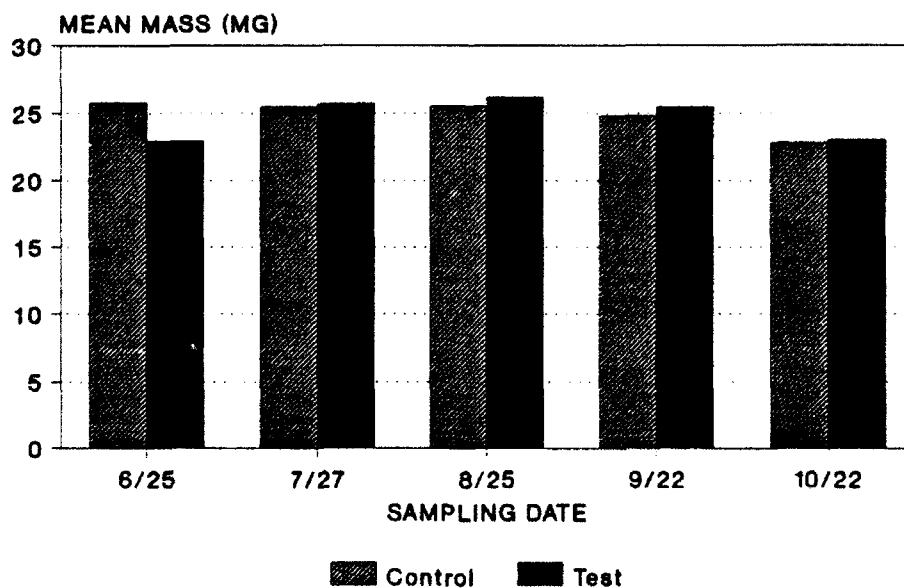


Fig. 41. Mean mass of cocoons produced by *A. tuberculata* of Test and Fire Tower provenance in 1992.

C.4. Clitellate proportions

Following the removal of A. tuberculata from EM field influence (Test provenance series incubated in Control in 1991), a larger proportion of adults became clitellate (with respect to those incubated in Test), significantly so in September and October (Fig. 42). The trend continued through July 1992, then was reversed in the last three months of the season, when clitellate numbers gradually declined.

Omitting the first sampling date (the redistribution scheme had not yet taken effect), ANOVA of 1992 data yielded high significance levels for all factors (Table 24). The date x site interaction in particular quantified the dissimilar patterns of reproduction in Test and Control bags during the first and second half of the season (Fig. 42). Date-specific tests confirmed that significantly higher Control means in May, June and July ($P < 0.01$ or better) were followed in September by higher mean clitellate proportions in Test bags ($P < 0.001$).

Table 24. Results of ANOVA of proportions of A. tuberculata in the clitellate state, Test Provenance series, 1992.

Source	SS	DF	MS	F-ratio	P
Date	0.5092	5	0.1018	32.26	0.0000
Site	0.1422	1	0.1422	45.05	0.0000
Date x site	0.4323	5	0.0865	27.39	0.0000
Error	0.1515	48	0.0032		

TEST PROVENANCE 1991-92

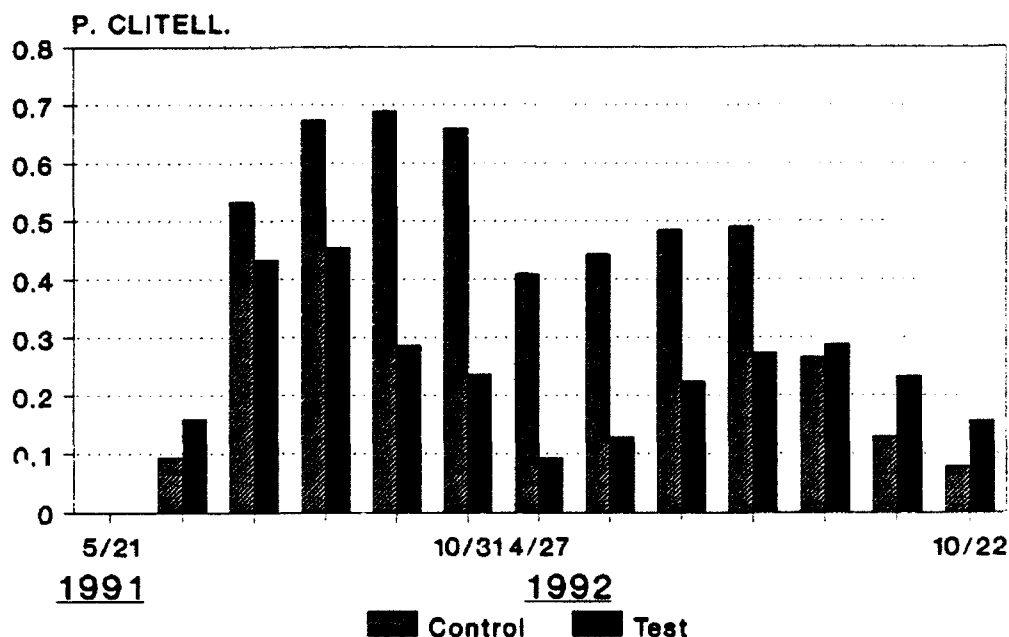


Fig. 42. Mean proportion clitellate among *A. tuberculata* of Test site provenance, incubated in Test and Control in 1991-92.

Among earthworms newly exposed to EM fields (Fire Tower series incubated in the Test site in 1992), up to 88% became reproductive, clitellate proportions in Control bags slightly exceeding those in Test bags only in October (Fig. 43).

Unable to normalize these data, we used Kruskal-Wallis non-parametric Anova to test for between-site differences. Results ($P = 0.12$) showed that mean clitellate proportions in Test did not differ significantly from those in Control.

FIRE TOWER PROVENANCE 1992

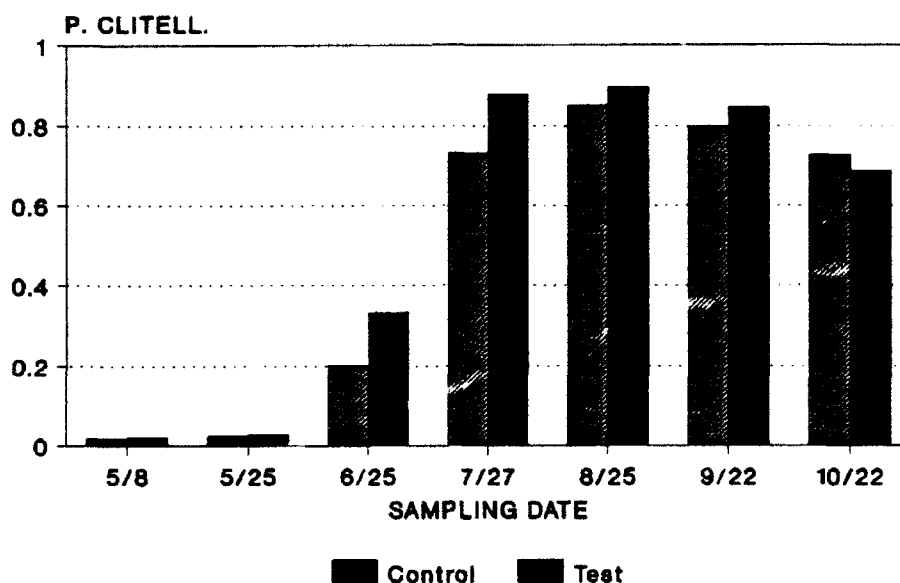


Fig. 43. Mean proportion clitellate among A. tuberculata of Fire Tower site provenance, incubated in Test and Control in 1992.

C.5. Cocoon numbers

Mean numbers of cocoons produced by A. tuberculata of Test provenance, incubated in Control bags, generally exceeded those produced by worms incubated in Test (Fig. 44). In 1991, means differed significantly only in late October ($P < 0.05$). Date-specific tests of 1992 data showed that means differed on three occasions: in June, worms incubated in Control out-produced those in Test ($P < 0.001$), while the reverse was observed in September and October ($P < 0.001$). Clearly, cocoon numbers were merely a reflection of the relative numbers of clitellates present in Test and Control bags (Fig. 42).

TEST PROVENANCE, 1991-92

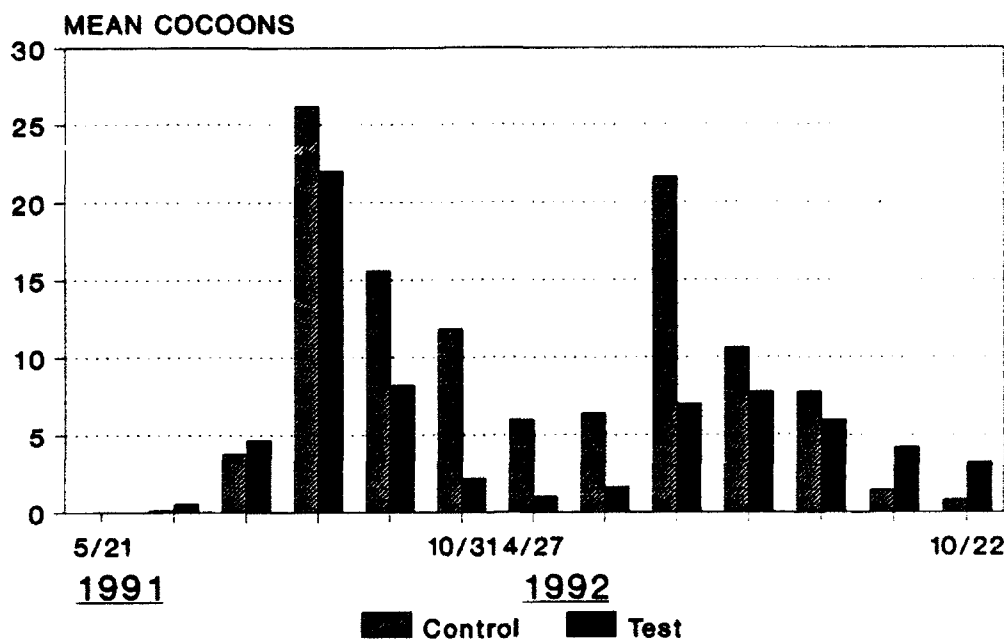


Fig. 44. Mean numbers of cocoons produced by A. tuberculata of Test provenance, 1991-1992.

Fire Tower provenance A. tuberculata newly exposed to EM fields produced significantly more cocoons than those incubated in Control (July, August and September, $P < 0.01$ or better) (Fig. 45). In fact, differences between Test and Control data with respect to cocoon numbers (Fig. 45) appeared more striking than differences with respect to clitellate proportions (Fig. 43).

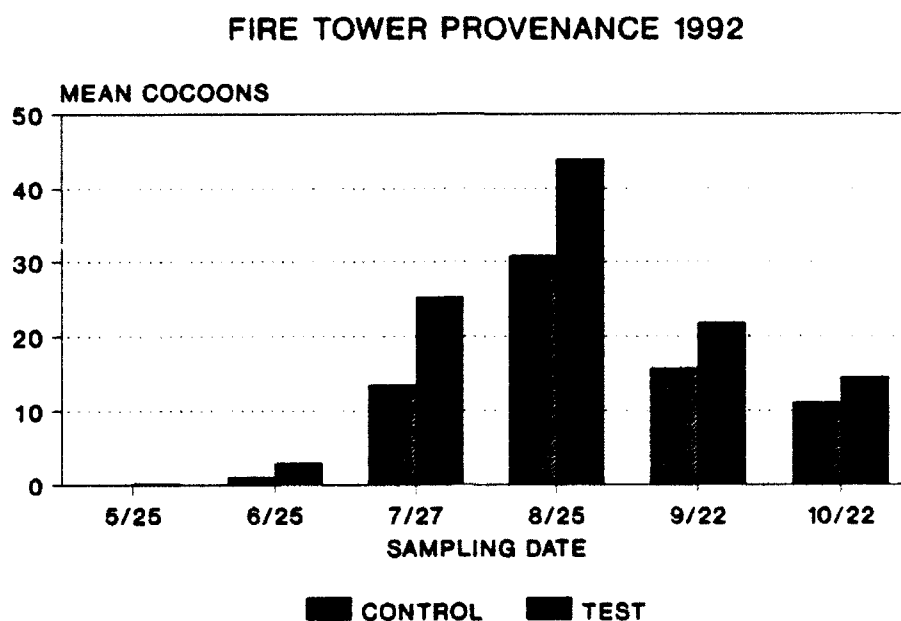


Fig. 45. Mean number of cocoons produced by A. tuberculata of Fire Tower provenance, incubated in Test and Control in 1992.

C.6. Cocoon production rates

Ratios of cocoons/clitellates can be expressed in several ways. For field-derived data, we described two simple methods based on numbers of cocoons and reproductive adults in section V.2. These methods (1 and 2) were also applied to experimental wormbag data.

In the case of confined A. tuberculata, however, mass estimates for earthworms and for the cocoons produced by them during each incubation period were also available. They were used to estimate production rates by two methods (Methods 3 and 4) analogous to those based on numbers. Resulting values are meaningful in terms of "reproductive effort", given that production of cocoons represents

an expenditure of energy.

Method 3 relates total mass of cocoons on date_i to "average" total mass of clitellates $[(\text{mass}_{\text{date } i} + \text{mass}_{\text{date } i-1}) / 2]$, and is thus a mass-based equivalent of Method 1.

Method 4 relates total mass of cocoons on date_i to total mass of clitellates on date_i, and represents a mass-based equivalent of Method 2.

Two-factorial ANOVA was possible for all estimates of cocoon production rates. Results pertaining to Test provenance A. tuberculata (1992 data) are listed in Table 25; the corresponding date-specific mean rates are illustrated in Figs. 46 and 47.

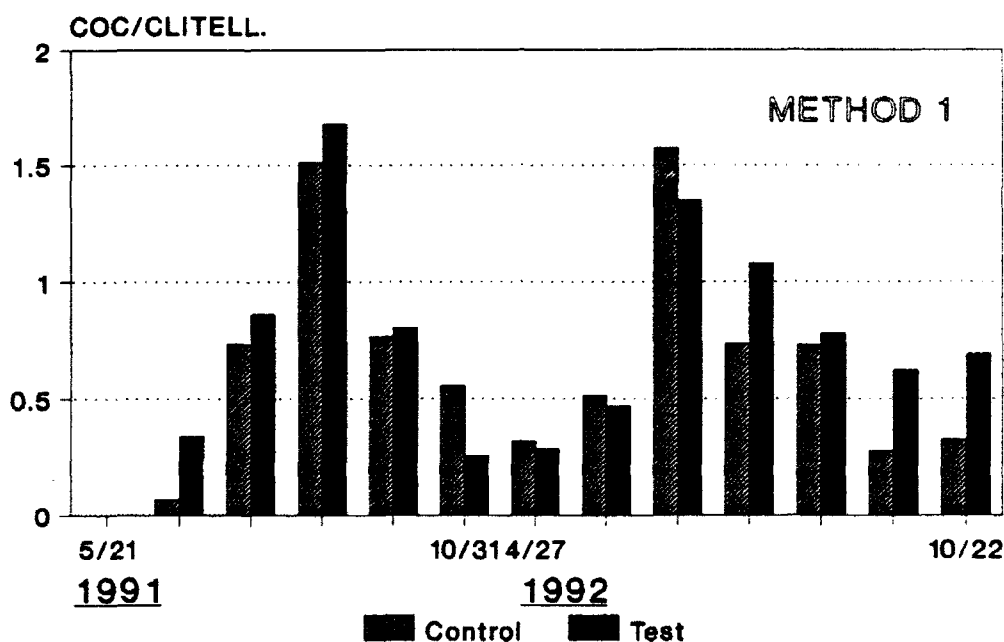
As expected, date effects were significant in all cases, due to effects of seasonal temperature changes. Site effects were significant for Methods 1 and 3, but not 2 and 4 (which are derived from cocoons and clitellates on date_i, and thus disregard the potential influx or decrease in clitellate numbers during each incubation period). Significant date x site interactions in all four cases reflect temporal differences in reproductive patterns in Test and Control (Figs. 46 and 47).

With the single exception of October, mean ratios observed in 1991 were higher for worms incubated in the Test site (Fig. 46). Date-specific tests, however, did not show the means to differ significantly between sites (results were affected by high variances due to the experimental design, which did not involve redistribution of earthworm groups; ref. section 4.A.).

Table 25. Results of ANOVA of 1992 cocoon production rates by Test provenance A. tuberculata, incubated in Test and Control sites (for explanation of Methods, see text).

Source	SS	DF	MS	F-ratio	P
Method 1					
Date	7.4669	5	1.4934	27.24	0.0000
Site	0.2858	1	0.2858	5.21	0.0269
Date x site	0.7647	5	0.1529	2.79	0.0273
Error	2.6317	48	0.0548		
Method 2					
Date	4.4605	5	0.8921	10.91	0.0000
Site	0.0040	1	0.0040	0.05	0.8263
Date x site	1.6098	5	0.3220	3.94	0.0045
Error	3.9242	48	0.0818		
Method 3					
Date	0.0038	5	0.0008	23.19	0.0000
Site	0.0002	1	0.0002	7.51	0.0086
Date x site	0.0005	5	0.0001	2.96	0.0210
Error	0.0016	48	0.0000		
Method 4					
Date	0.0028	5	0.0006	10.43	0.0000
Site	0.0000	1	0.0000	0.42	0.5178
Date x site	0.0012	5	0.0002	4.64	0.0016
Error	0.0026	48	0.0001		

TEST PROVENANCE 1991-92



TEST PROVENANCE 1991-92

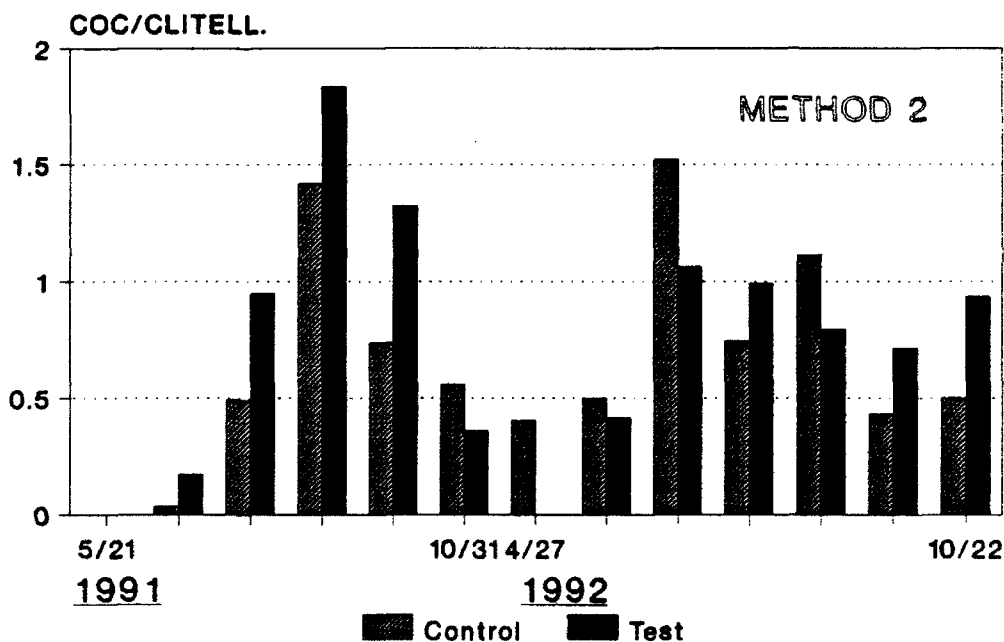


Fig. 46. Mean cocoon production rates (based on total numbers) of *A. tuberculata*, Test provenance series, 1991-92 (sampling intervals of 1 month); for explanation of Methods, see text.

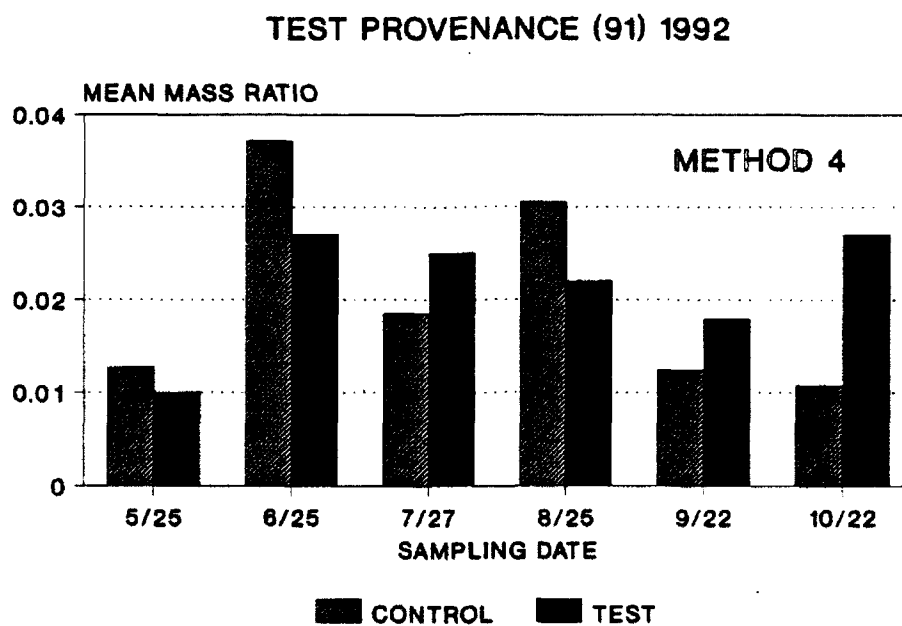
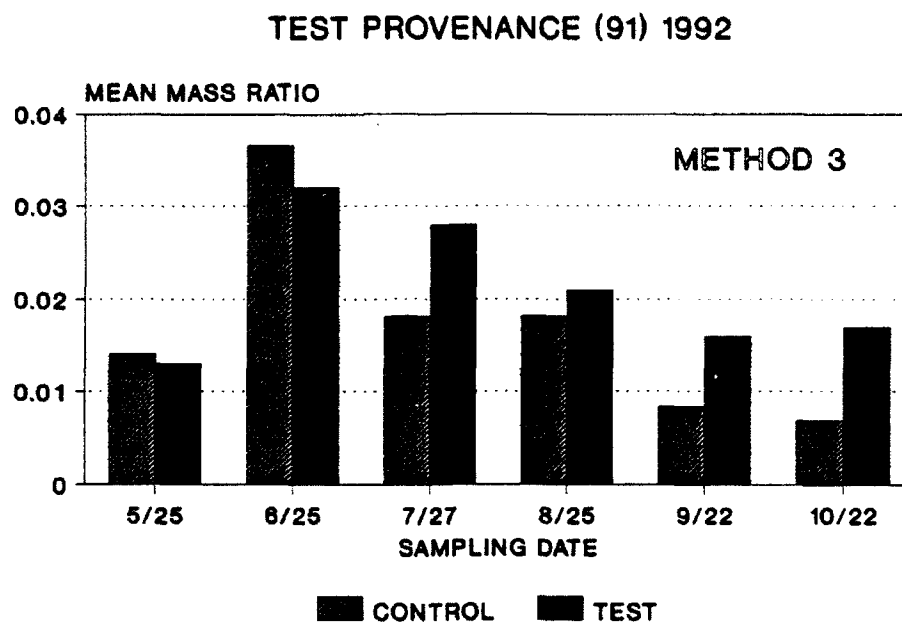


Fig. 47. Mean cocoon production rates (based on total mass) of Test provenance *A. tuberculata* during 1992 (sampling intervals of 1 month); for explanation of Methods, see text.

Fire Tower provenance A. tuberculata had never been exposed to EM fields of any magnitude. Their response to incubation in Test bags in terms of cocoon production rates (by whatever method they were calculated) was unequivocal.

Site effects were highly significant (Table 26), production rates in Test exceeding those in Control on all dates (Figs. 48 and 49). Absence of date x site interaction indicated that seasonal trends (determined by temperature) were strongly parallel in the two sites; and that exposure to EM fields was the most likely cause underlying increased rates of cocoon production in the Test site.

Table 26. Results of ANOVA of cocoon production rates, Fire Tower provenance A. tuberculata, 1992; for explanation of Methods, see text.

Source	MS	DF	SS	F-ratio	P
Method 1					
Date	16.6889	4	4.1722	79.29	0.0000
Site	2.3642	1	2.3642	44.93	0.0000
Date x site	0.1839	4	0.0460	0.87	0.4830
Error	4.7357	90	0.0526		
Method 2					
Date	16.7977	4	4.1994	124.56	0.0000
Site	2.3389	1	2.3389	69.38	0.0000
Date x site	0.2895	4	0.0724	2.15	0.0815
Error	3.0342	90	0.0337		
Method 3					
Date	0.0095	4	0.0024	118.98	0.0000
Site	0.0008	1	0.0008	37.96	0.0000
Date x site	0.0001	4	0.0000	0.64	0.6351
Error	0.0018	90	0.0000		
Method 4					
Date	0.0121	4	0.0030	192.10	0.0000
Site	0.0009	1	0.0009	54.38	0.0000
Date x site	0.0001	4	0.0000	1.84	0.1289
Error	0.0014	90	0.0000		

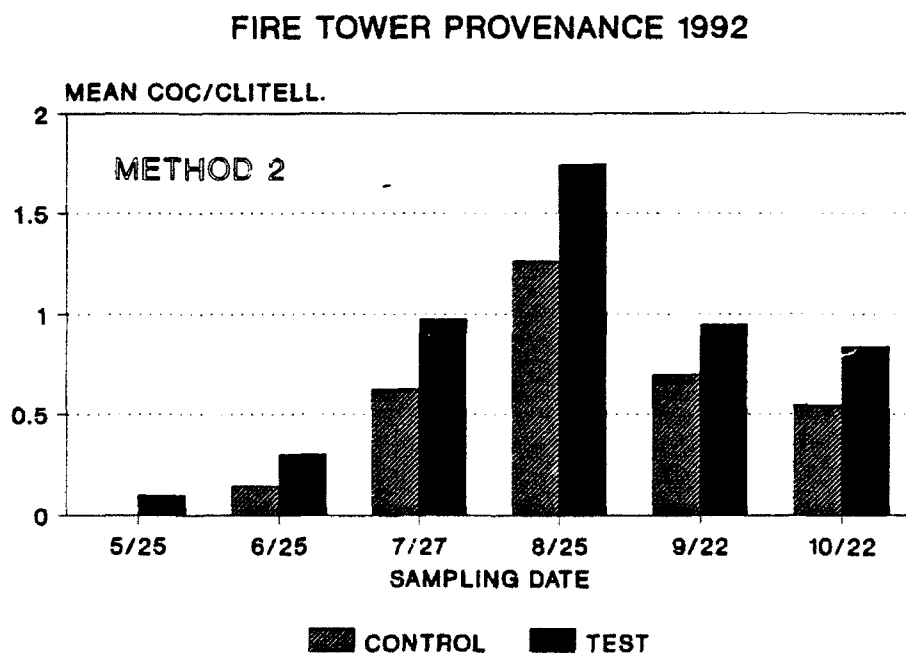
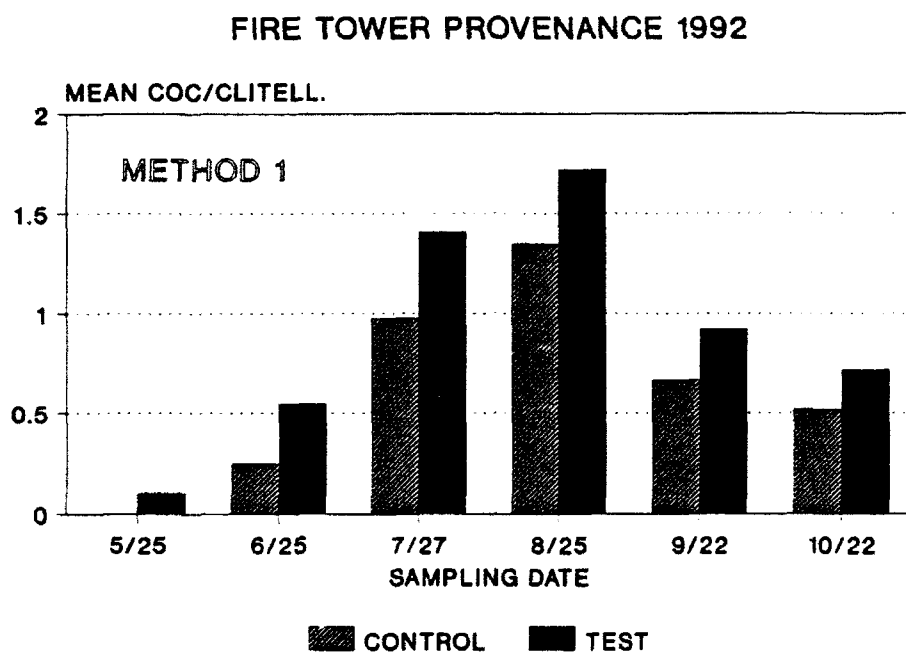


Fig. 48. Mean cocoon production rates (based on total numbers) by *A. tuberculata* of Fire Tower provenance, incubated in Test and Control in 1992; for explanation of Methods, see text.

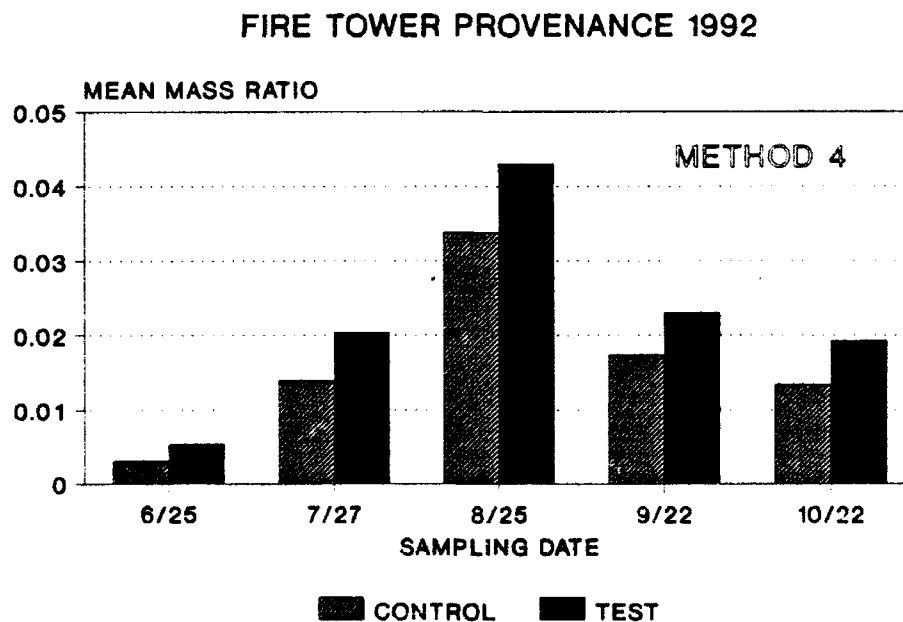
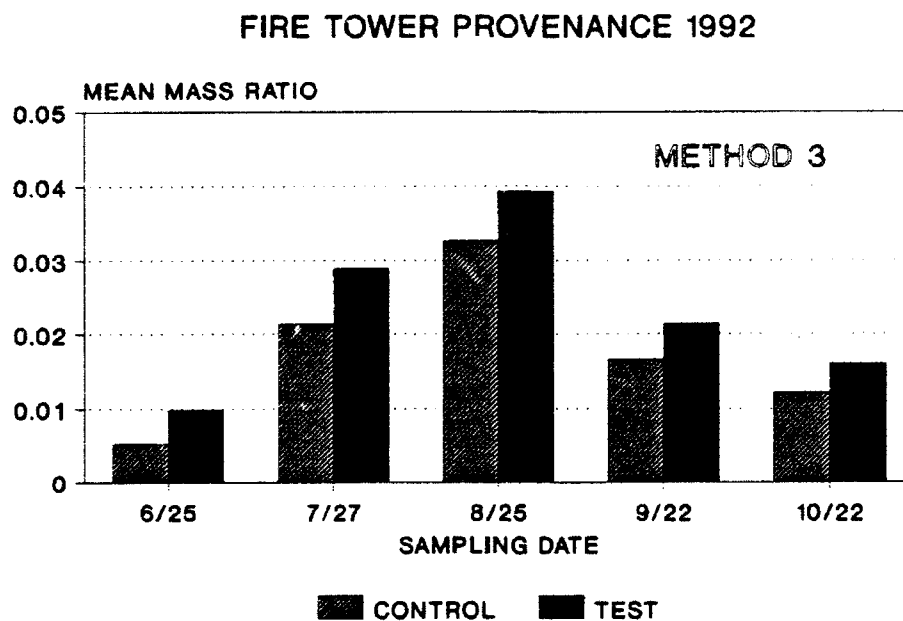


Fig. 49. Mean cocoon production rates (based on total mass) by Fire Tower provenance *A. tuberculata*, incubated in Test and Control in 1992; for explanation of Methods, see text.

D. Conclusions

Apparent contradictions between field population data and those derived from earthworm isolation experiments can be tentatively explained if increased cocoon production rates are viewed as the primary effect of exposure to EM fields. Higher rates imply greater expenditure of energy (in the form of cocoons). It is plausible that adults leave the pool of reproductive individuals sooner (i.e., become post-reproductive after a shorter time period) than they would without EM exposure; the total number of clitellates present in the population at any given time would thereby be reduced.

The data also indicate that effects on cocoon production rates are immediate as well as long-lasting, but that secondary effects on clitellate adult numbers exhibit a time delay (in the Fire Tower incubation series, these effects have yet to take place). Extrapolation from experimental to field data is further complicated by at least three factors:

a.) Moisture levels in mesh bags are less variable and less stressful than they may be in the natural habitat at certain times of the year.

b.) The substrate (soil) used in mesh bags appears to be more conducive to maximum growth of A. tuberculata than the locally variable natural soil. Evidence for this surmise is found in body mass data: field-collected adults very rarely weigh > 1000 mg, average mass of clitellates typically being 400 to 800 mg. In view of energy-expenditure considerations, secondary effects (onset of

a post-reproductive phase) may occur sooner in smaller animals under actual field conditions.

c.) On the average, EM field intensities are somewhat higher in the natural habitat than in mesh bags, although they are variable in both types of environment. IF a dose-response relationship exists with respect to magnitude and speed with which EM influence takes effect, then comparison between field and experimental data must be done with caution.

We believe that continued monitoring of field-incubated earthworms, now entering their second year of EM exposure, will clarify current results. In addition, we propose to repeat the "Test Provenance" series, using earthworms collected in Test (now in their fifth year of exposure to EM fields) and incubated in Test and Control sites.

5. Lumbricus rubellus

We have been unable to develop predictive models for reproductive activity of Lumbricus rubellus during pre-operational years, since none of the environmental variables tested yielded adequate coefficients of determination. The relative imperviousness of the species to climatic fluctuations, coupled with typically low adult numbers in the population, introduce a great deal of random error into data analyses. Below, we present large-scale summaries which characterize the species' life cycle and its variations during 1988-92; they indicate that efforts toward rigorous analyses should be pursued.

A. Reproduction

Mean annual clitellate proportions (= percent of all adults in the clitellate state) were lower in 1990 through 1992 than in any preceding year; in the first year of antenna operation (1989) they were approximately equal to those obtained for 1984 and 1987 (Table 27).

Even though predictive relationships could not be established, there appears to be a pattern in these estimates: during the years following antenna activation, clitellate proportions did not differ significantly from those observed in partial or severe pre-operational drought years, despite favorable moisture conditions (Table 3, section II). With respect to 1984 and 1987 (relatively high-moisture years), 1991 in particular yielded significantly fewer reproductive adults ($P < 0.01$); i.e., we continue to observe

that L. rubellus, in moist operational years, reproduces at rates equivalent to those observed in dry pre-operational years.

Table 27. Mean percent \pm SD Lumbricus rubellus (of all adults) in the clitellate state, 1984-1992 (N dates in parentheses).

YEAR	1984	1985	1986	1987	1988	1989	1990	1991	1992
Mean %	85.3	69.0	69.1	79.1	69.3	84.2	60.9	56.9	64.4
\pm SD	14.3	26.3	17.4	22.8	21.4	15.1	19.4	24.4	18.5
(N)	(12)	(13)	(13)	(10)	(12)	(13)	(13)	(13)	(12)

Abundance of cocoons exhibited some modulation by severe drought (1988) or high moisture conditions (1987) during pre-operational years (Fig. 50). Among operational years, 1991 was distinguished by low numbers of total adults and clitellates (Fig. 51), traceable to low cocoon production in 1988. Numbers of adults, clitellates, and cocoons reached an all-time high in 1992 (Figs. 50-51), a direct result of high survival rates of the 1990 cohort of hatchlings (ref. section B. below).

There is little doubt that edaphic conditions influence the level of reproduction in L. rubellus to some degree. Whether increased variability during operational years was solely due to environmental factors, or to potential ELF effects in interaction with these factors, remains unresolved at this time. If EM fields affect L. rubellus as they do A. tuberculata, it is conceivable that reproduction was initially stimulated in 1989, followed by depression of clitellate proportions in subsequent years. Analyses

of cocoon production rates, a parameter which proved crucial in A. tuberculata, are currently underway.

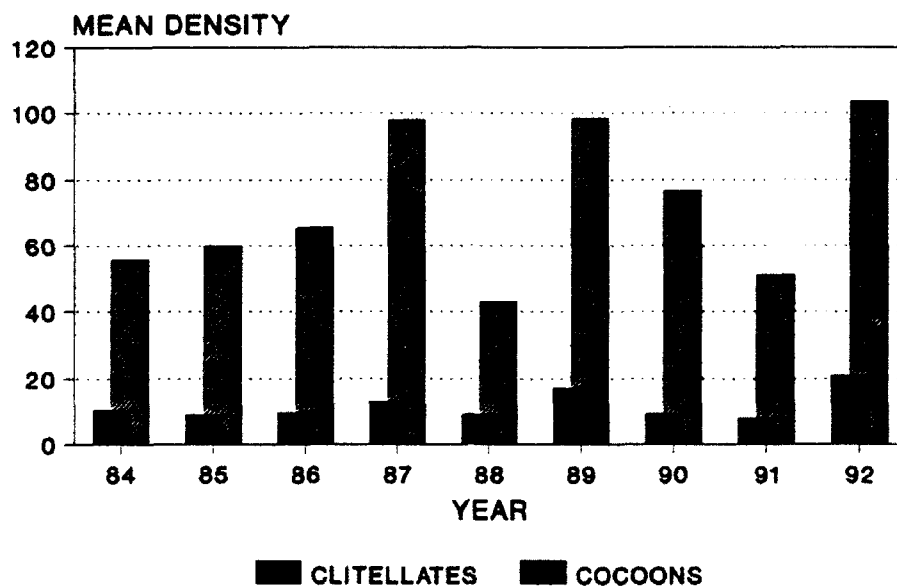


Fig. 50. Mean annual density of clitellate adults and new cocoons of Lumbricus rubellus, 1984-1992.

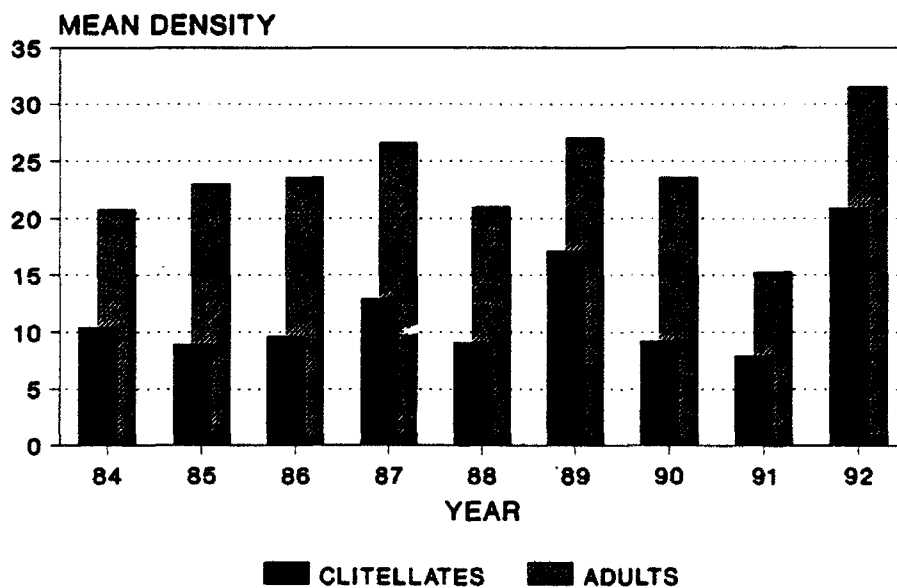


Fig. 51. Mean annual density of total adults and clitellates of Lumbricus rubellus, 1984-1992.

B. Population structure

Patterns of growth and maturation, detailed in earlier reports, can be summarized as follows: fully developed cocoons accumulate in the fall, and hatchlings emerge in very early spring of the following year. We estimated that at least two years of growth are required to reach adulthood. During pre-ELF years, growth patterns (measured by abundance of individuals in successive weight classes) was often obscured by high mortality during drought years. Beginning in 1989, however, we were able to document the development of a single cohort.

High cocoon numbers in 1989 resulted in a hatchling peak in May of 1990 (Fig. 52). Most of these juveniles reached class 3 size in October 1990, and grew to large immatures (classes 4 and 5) during 1991. In 1992, a significant increase in large immatures and adults, stemming from 1989 cocoons, was observed (Fig. 52). Growth from hatchling to adulthood thus requires approximately 2 to 2.5 years in L. rubellus.

Population structure expressed as immature/adult ratios was found to be more variable during operational years than during 1984-88 (Fig. 53).

Mean annual immature proportions were approximately equal during all five pre-ELF years. The severe 1988 drought is believed responsible for low immature numbers in 1989, due to increased mortality as well as low cocoon production. High cocoon production in 1989, coupled with high survival rates of the resulting 1990 cohort of hatchlings, produced unusually high immature proportions

in 1990 and 1991 (Fig. 53). Low immature proportions in 1992 are traceable to two factors: low cocoon numbers in 1991 (= low numbers of hatchlings in 1992) and high numbers of adults stemming from the May 1990 cohort of recruits (Fig. 52). It appears that protracted periods (2 or more years) of favorable moisture conditions are likely to produce greater year-to-year variability in immature/adult ratios, mainly through high survival rates of a given cohort.

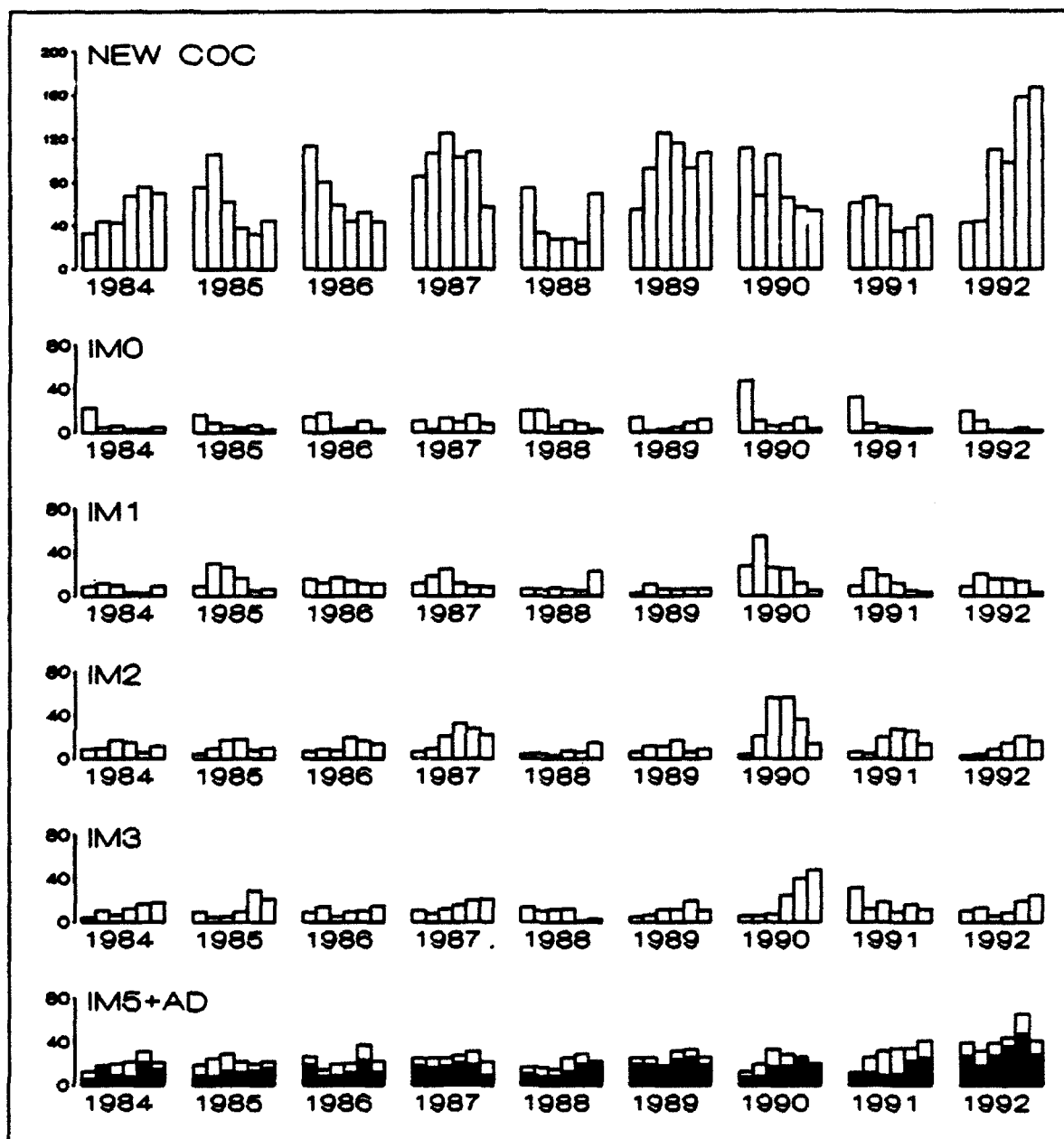


Fig. 52. Mean monthly densities (May through October) of new cocoons, selected immature classes and adults of *L. rubellus*, 1984-1992. IM 0 = hatchlings; IM 1 = 13 - 26 mg, IM 2 = 26.1 - 52 mg, IM 3 = 52.1 - 104 mg, IM 5 = >208 mg. Black portion of bars in last row = adult densities.

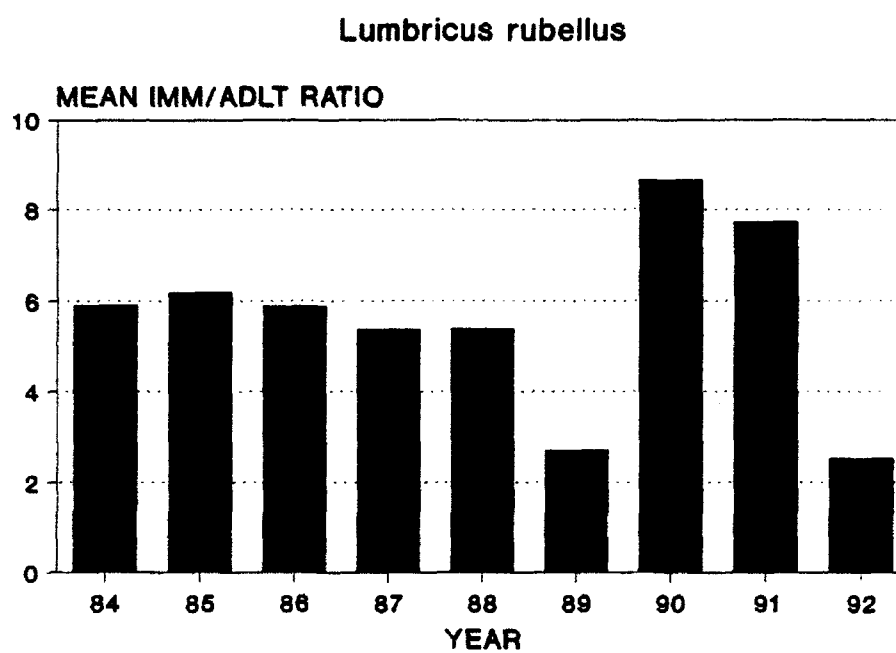


Fig. 53. Mean annual immature/adult ratios of L. rubellus, 1984-1992.

VI. LITTER INPUTS AND DECOMPOSITION

1. Litter inputs

Consistent with previous years, maximum leaf-fall in 1992 occurred during the last week of September and the first week of October in both sites (Fig. 54). Total inputs were again similar to those recorded for the preceding 9 years (Table 28), and BACI analyses of seasonal litter inputs in Test and Control confirmed that no effects of antenna operation were detectable (Table 1).

Table 28. Annual litter inputs (g dry/m²), 1983-1992, by the dominant Acer saccharum and by all species together in Test and Control.

		1983	1984	1985	1986	1987	1988	1989	1990	1991	1992
Maple	T:	189	177	203	176	161	191	180	169	167	159
	C:	221	179	199	189	180	198	162	172	187	176
Total	T:	278	259	286	252	231	276	269	246	261	250
	C:	305	264	289	284	275	301	258	261	296	277

2. Decomposition

Litterbag series IV (initiated November 1990) was sampled for the last time in May of 1992, after a second winter in the field. Series V (initiated November 1991) was sampled at monthly intervals throughout 1992, and a final sample will be obtained in May 1993. Series VI was placed in the field in November 1992, to be retrieved during 1993.

Decomposition data for 1992 are not available at this time because ashing of samples was interrupted by equipment failure. Data will become available by early summer. A preliminary assessment, however, indicates that ELF effects are not likely to have occurred in 1990-1992.

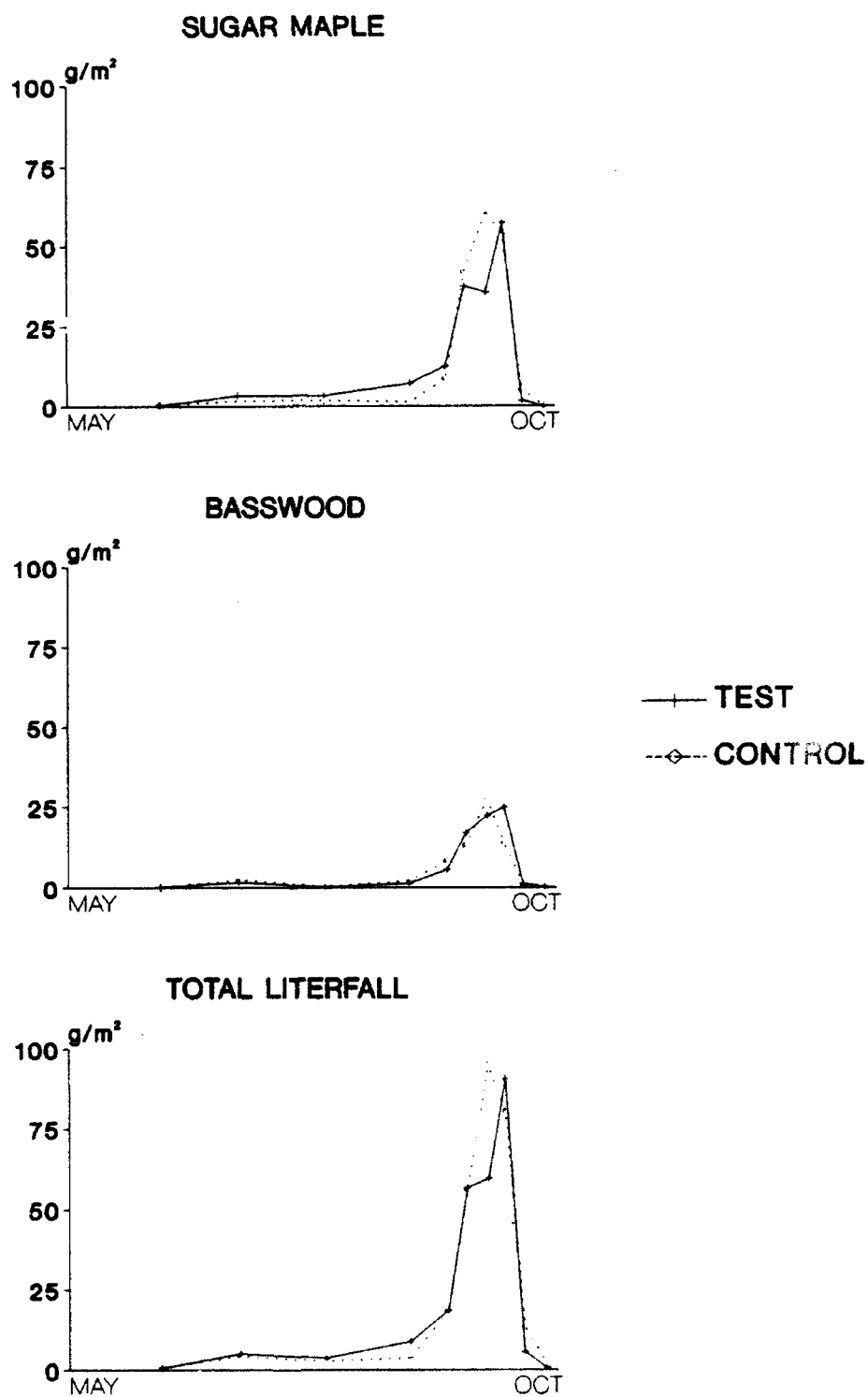


Fig. 54. Litter inputs (g dry/m^2) in Test and Control in 1992.

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BIOLOGICAL STUDIES ON POLLINATING INSECTS: MEGACHILID BEES

Annual Report 1992

Karen Strickler
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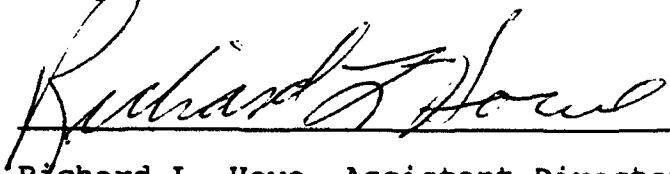

Richard L. Howe, Assistant Director
Contract and Grant Administration

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GLOSSARY AND LIST OF ACRONYMS

C5: Camp 5 control site

CATMOD: Categorical data modeling procedure in SAS.

CL: County Line control site

ELF: Extremely Low Frequency

EM: Electromagnetic

Exp: Variable indicating whether the data were from an experimental or a control area.

Exp*Year: Interaction effect of the Exp and year variables in the GLM, ANOVA, or CATMOD model.

Expected Sex: The actual or predicted sex of the bee offspring in a cell. Predicted sex is based on the order of the cell in the nest, and the presence of at least one cell of known sex. Females are found in the innermost cells, males in the outermost cells (see p. 14,16).

F1: Ford 1 (north Ford) experimental site

F2: Ford 2 (south Ford) experimental site

GLM: General Linear Modeling procedure in SAS.

LO: A round leaf piece used to cap a cell or plug a nest. Occasionally an LO is found at the base of a cell or is part of the construction of a cell lining, along with LRs. The bee carries an LO in her mandibles.

LR: An elongate, oblong leaf piece used to line a cell. The bee carries an LR rolled between her legs.

Measurer: variable indicating the person who observed or measured data.

Ow Site: Overwintering site

Primary sex ratio: The sex ratio that would have been produced if all cells had yielded an offspring.

SAS; Statistical software package on the VAX computer, used in analysis of data.

Season (early vs. late): Nests were classified as "early season" if they were begun on or before the date on which half of the nests of that species were begun during that year.

Nests begun on later dates were classified as "late season" nests.

Secondary sex ratio: The ratio of male to female adult and pupal offspring, which could be sexed with certainty.

Site [exp]: Site variable nested in experimental areas.

Trip Rank: The number of LO leaves already collected by a bee, including the current LO, in a series of LO trips to cap a cell. Usually the duration of the first 5 such trips are recorded for a given cell cap. These LO trip durations are given Trip Ranks of 1,2,3,4, and 5 respectively.

Yr: Year

I ABSTRACT

High voltage transmission lines and magnetic fields have been shown to affect honeybee reproduction, survival, orientation, and nest structure. ELF EM fields could have similar effects on native megachilid bees.

Two species in the genus *Megachile* have been abundant in artificial nests at experimental and control sites in Dickinson and Iron Counties in Michigan. Data on their nest architecture, nest activity, and emergence/mortality have been collected since 1983. Five hypotheses concerning the possible effects of ELF EM fields are considered using these data. The ELF ANTENNA has been at 100% power since the summer of 1989. Exposure to ELF EM fields could be reflected in data for 1989 - 1991.

Our hypotheses have involved monitoring changes in cell length, number of cells per nest, length of nest plug, number of leaves per cell, orientation of nest entrances, time to collect a round leaf piece to cap a cell, and overwintering mortality. Our statistical analyses have been improved this year. Thus far during operational years we have not detected significant changes in cells per nest, nest plug lengths, nest orientation, or time to collect a leaf at experimental areas that could be attributed to ELF EM fields. *M. relativa* cells may be about 0.1mm larger at experimental sites as a result of ELF EM fields, and *M. inermis* may add an average of one additional leaf per cell at experimental areas since the antenna became operational. These increases are deemed unlikely to impact the bee population.

This year we have added a statistical test for the effects of ELF EM fields on sex ratio in *M. relativa*, but did not find any significant changes associated with the antenna.

M. inermis cells and nests with prepupal mortality at experimental sites have increased more since the antenna became operational than have cells and nests with prepupal mortality at control sites. When nests from one experimental site are moved to a control site for the winter, mortality is reduced to the level of the control site. These results suggest that ELF EM fields increase overwintering mortality. Overwintering data for 1992 nests will be added to our analysis this summer. We will be interested to see if increased mortality at experimental sites persists.

II INTRODUCTION

Project Rationale and Overall Objectives.

High voltage transmission lines and fluctuations in the earth's magnetic field have been reported to affect honeybees (Greenberg et al. 1981; Gould 1980). In addition, honeybees have been shown to have an organ in the abdomen consisting of magnetite particles that could be used to detect the earth's magnetic field and thus could be used as a compass in orientation (Gould et al. 1978). This organ appears to be involved in the detection by foraging honeybees of localized magnetic anomalies associated with nectar rewards (Walker and Bitterman, 1989; Kirschvink and Kirschvink, 1991). Honeybees appear to use the earth's magnetic field as a reference system for orientation based on polarized light, and the presence of an artificial magnetic field causes a positive deviation in the angle of the waggle dance for bees orienting their dance on a horizontal hive where skylight but not the sun is visible (Leucht and Martin, 1990). Because such effects of electric and magnetic fields have been demonstrated, it is possible that ELF EM fields may alter a bee's ability to orient or may otherwise affect its behavior.

Honeybees, however, are rare in the state forest where the Michigan ELF antenna is located (personal observation), and are unable to overwinter in the harsh climate of Michigan's Upper Peninsula (Fischer, 1983 Annual Report). Therefore, native bees are a better choice for ecological studies of the resident bee fauna. Native bees are particularly important in ecological communities such as those in the vicinity of the ELF antenna because they are pollinators of flowering plants, and are therefore important to the reproductive success of these plants.

With the exception of bumblebees and some halictids, native bees are solitary, meaning that each female constructs and provisions her own nest rather than having a special queen caste responsible for reproduction. Solitary bees have several advantages for ecological studies. As "mass provisioners", they create a discrete cell for each offspring, and fill it with a provision mass of pollen and nectar prior to laying the egg. The bee does not add more provisions after the egg is laid. A series of such cells, each with a provision mass and egg, are created in succession by each female. The provisions that go into each cell are a direct measure of parental investment in an offspring (Strickler 1979; Cowan 1981; Johnson 1983; Danforth 1990). The size of the adult bee that emerges from each cell is correlated with the amount of provisions provided it, and with the size of the cell in which the larva

develops (Krombein 1967; Klostermeyer et al. 1973; Trivers and Hare 1976; Alcock 1979; Torchio and Tepedino 1980; Johnson 1983; Danforth 1990). However, there is a tradeoff between the investment per offspring and the rate at which offspring are produced. The more the bee invests per offspring (ie, the larger the offspring), the fewer offspring she will produce. If bees are disoriented, agitated, or slower at foraging, they may invest less per offspring, produce fewer offspring per unit time, or both. Solitary bees are unusual in having this direct relationship between parental investment per offspring, adult size, and reproductive output.

The nesting biology of some species of solitary bees in the family Megachilidae is especially easy to study because they accept artificial nests placed in the field. These bees typically nest in abandoned beetle bores in dead logs. "Trap nests" of drilled blocks of wood are also used by bees as nest sites. Such artificial nests can be placed in habitats where bees are expected to nest, in order to increase the sample of nests available for study, and to standardize such characteristics of the nest as bore depth and diameter (Krombein, 1967). Trap nests are used in the management of the Alfalfa Leafcutting Bee, *Megachile rotundata*, for pollination of alfalfa (Stephen, 1962, 1981; Bohart and Knowlton, 1964; Johansen et al., 1969; Bohart, 1972; Gerber and Klostermeyer, 1972; Hobbs, 1972; Baird and Bitner, 1991), and the Blue Orchard Bee, *Osmia lignaria* for the pollination of fruit trees (Torchio 1981a,b; 1982a,b,c; 1984a,b; 1985). Thus there is an extensive (though largely unreviewed) literature on megachilid biology. Literature relevant to the ELF project is discussed throughout this report.

Although the effects of electromagnetic fields on solitary bees had not been studied previous to the ELF project, research on the effects of high tension wires and magnetic fields on honeybees suggested working hypotheses on which to base our analyses of megachilid nesting biology. Of possible relevance to megachilid behavior are an alleged greater tendency for dispersal, and greater levels of activity (Wellenstein, 1973), as well as reduced reproductive output, lower overwintering survival, and modifications of nest structure (Greenberg et al., 1981a,b) when colonies were exposed to electromagnetic fields from high voltage transmission lines. Disturbance of colonies under transmission lines can be attributed to electric shock from induced hive currents, especially under wet conditions (Bindokas et al., 1988). Although induced currents are less likely in trap nests than in honeybee hives, the possibility of stress or disturbance from electromagnetic fields should be appraised. In addition, disorientation due to fluctuations in ELF magnetic fields is possible if megachilids share the honeybee's ability to detect magnetic fields. (Gould et al., 1978, 1980; Gould 1980; Tomlinson et al. 1981; Walker and

Bitterman, 1989; Kirschvink and Kirschvink, 1991). No data exist on the ability of megachilids to detect magnetic fields.

Nesting Biology of Megachilid Bees

A decision to restrict our study to two species of leaf-cutting bees, *Megachile (Megachile) relativa* Cresson and *Megachile (Megachile) inermis* Provancher, was made in the fall of 1986 (1986 Annual Report). *M. inermis* and *M. relativa* have similar nest architecture in that both line their cells with pieces of cut leaves. However, the two species differ in size, and may therefore partition their time and the space in their nests differently. Aspects of the biology of both species have been described generally for populations in Wisconsin and Canada (Medler, 1958; Medler and Koerber, 1958; Stephen, 1955, 1956; Longair, 1981).

The general structure of the nests of the two species is depicted in Fig. 1. The bee may leave some space at the base of the nest (the basal space) unoccupied by cells for offspring. She may then cut and bring to the nest a few round pieces of leaf that are added one at a time to form the base of the first cell. Next she cuts and brings to the nest several elongate pieces of leaf (LRs) in succession. These are used to line a tube- or cup-shaped cell that is slightly longer than her body. Next she makes a series of pollen and nectar foraging trips to fill the cell with the discrete provision mass that will be the larva's food supply. When provisioning is complete, the female lays an egg. Fertilized eggs become females while unfertilized eggs become males. The female has voluntary control over fertilization and thus the sex of the offspring in each cell (Klostermeyer and Gerber, 1970). After laying the egg, she cuts more leaves, this time round in shape (LOs), to cap the cell. Sometimes she adds chewed leaves, dirt, or bits of wood to separate the cells. Next she cuts more elongate leaves for the second cell, and repeats the process. Thus a linear series of cells is constructed in the nest bore. Typically, the cells at the base of the nest are more likely to contain females and the cells near the entrance are more likely to contain males (Krombein, 1967). Since females are usually larger than males in these bees, cells at the base of the nest tend to be larger than cells near the entrance. When she has completed the last cell that she is going to put in the nest, she constructs a series of plugs of round leaves, chewed leaves, dirt, chewed wood, and possibly other material. *M. relativa* frequently includes empty "vestibular" spaces between segments of plug. *M. inermis* and some *M. relativa* create one long mass of plug material after completing the reproductive cells. In nests of both species there is usually a space between the outermost plug and the opening of the nest, called an "indentation".

Each female may construct several such nests over her life time. The adult life span is no more than one season; adults do not overwinter. Some nests are abandoned before they are finished because the bee has died, or for other unknown reasons. Some incomplete nests may be usurped by other species of wasps and bees, which construct their own nests in the unused space of the trapnest.

Inside each cell the egg hatches, and the young larva feeds on the provisions prepared by its mother. Both *Megachile* species at our sites are univoltine (with a few exceptions; see Emergence Results), and both overwinter as prepupae. Pupation occurs in spring, and adults emerge soon after, in June and July at our study sites. A variety of parasites may emerge from the cell instead of the original bee. Oviposition by parasites of the genus *Coelioxys* (Megachilidae) often occurs while the cell is being provisioned, when the mother host bee is out of the nest on a pollen foraging trip, or on a round-leaf foraging trip just after laying her egg. Other parasites may lay their eggs in empty nests holes (*Anthrax* spp., Diptera: Bombyliidae) or in complete nests (chalcids; Hymenoptera: Chalcidoidea).

Hypotheses Tested

During the first four years of the project, 1983-1986, data on nest architecture, nest orientation, emergence/mortality and nest activity were collected. Based on these data, six tentative hypotheses concerning the effects of ELF EM fields on *Megachile* behavior were specified in the 1986 Annual Report. The initial hypotheses were modified in subsequent reports based on our ability to gather sufficient sample sizes to detect differences between experimental and control areas. The modified hypotheses are expressed in the following sections as null hypotheses, i.e., hypotheses of no difference between experimental and control areas, that we will try to disprove statistically. The "Rationale" sections explain the possible effects of ELF EM fields that may cause a rejection of the null hypothesis.

Hypotheses Involving Nest Architecture:

Hypothesis 1: The average length of cells for each offspring, and/or the average number of cells produced per nest is unchanged by exposure to ELF electromagnetic fields.

Rationale

Honeybee reproductive output decreased on exposure to high voltage transmission lines. Capped brood, which normally averaged 12,000 per hive, decreased to as low as no brood after 8 weeks of exposure (Greenberg, et al., 1981b). ELF EM fields

may have a similar effect on the number of cells produced by megachilids. Furthermore, ELF electromagnetic fields may affect cell size and nest architecture in various ways. For example, if bees are disoriented by the fields, they may gather resources (leaves, pollen) more slowly when exposed to the fields than when not exposed. As a result, they may produce new cells at a slower rate, or they may produce smaller cells.

Previous studies have found that the weight of offspring of the generalist megachilids, *Osmia lignaria* and *O. cornifrons*, is lower if their cells were produced late in the season rather than early in the season (Torchio and Tepedino, 1980; Sugiura and Maeta, 1989). These species also showed an increase in the proportion of male offspring (the smaller sex) produced late in the season. A reduction in offspring size late in the season is related to reduced foraging rates due to aging of the bee (Torchio and Tepedino, 1980, Tepedino and Torchio, 1982; Sugiura and Maeta, 1989). Similarly, ELF EM fields may slow the foraging of *M. relativa* and *M. inermis*, resulting in smaller bees produced in smaller cells. A size reduction could affect cells with offspring of both sexes, or it could reflect the production of a greater proportion of male offspring, since males are the smaller sex in both *Megachile* species. An additional complication is that female sizes decrease more than male sizes late in the season (Torchio and Tepedino, 1980). Thus we might expect female cells to be affected more than male cells by stresses from ELF EM fields.

In contrast to the generalist megachilids, the pollen specialist *Hoplitis anthocopoides* did not show a reduction in offspring weight late in the season, in spite of reduced foraging rates (Strickler, 1982). Rather, it was hypothesized that slower foraging rates led to fewer offspring per nest late in the season as compared with early in the season for this species. Similarly, *M. relativa* and *M. inermis* may produce fewer cells per nest in response to slow foraging rates due to ELF EM fields.

In testing hypothesis 1 we are interested in determining whether there are differences between experimental and control sites in cell lengths and number of cells per nest. Ideally, one hopes to find no differences between experimental and control sites, and between years, prior to the 1989 season when the ELF antenna was operational at full power. Then, if significant differences between experimental and control sites appear after the antenna is functioning at full power, we can attribute these differences to the effect of ELF EM fields.

Hypothesis 2: Bees exposed to ELF EM fields, and bees not exposed, will make nest plugs of the same thickness and will devote the same proportion of nest space to reproduction.

Rationale

Abnormal deposits of up to 48g of propolis were present at honeybee hive entrances under high voltage transmission lines, presumably in response to stress connected with electric fields at the nest entrance (Greenberg et al, 1981b). This suggests the possibility that megachilid bees will respond to disturbance from ELF EM fields by increasing the amount of nest "padding". This may be reflected in larger cells (tested in hypothesis 1) and/or increased nest plug length. More generally, there could be an increase in the nest space that does not include cells for offspring (ie. basal and vestibular spaces, nest plugs and indentations).

Hypothesis 3: The number of leaves used to line a cell is unchanged when bees are exposed to ELF EM fields.

Rationale

Bees may pad a cell with extra leaves as a result of stress due to electromagnetic fields, just as they may pad a nest with plug material. We can easily determine the number of elongate leaves used to line a cell by taking the cell apart after bee emergence and counting leaves.

Hypothesis 4: The relative acceptability of nests oriented in a NS direction vs. nests oriented in an EW direction does not change when bees are exposed to ELF EM fields.

Rationale

Honeybees may use the earth's magnetic field under special circumstances to orient their comb (reviewed in Gould, 1980). The fluctuating ELF magnetic fields could disturb any biases that megachilids normally have for nest orientation, or could cause greater acceptance of nests oriented in certain directions in order to reduce disturbance by the fields.

Hypotheses Involving Nest Activity

Hypothesis 5: The duration of round leaf (LO) foraging trips remains the same when bees are exposed to ELF EM fields.

Rationale

Honeybee activity, measured by honey production, allegedly doubled under high voltage electromagnetic fields in one study (Wellenstein, 1973). In contrast, colony weight, a measure of rate of honey accumulation and brood production, decreased by as much as half for colonies exposed to high voltage transmission lines in a different study (Greenberg et al., 1981b). In a third study, there were dose-related lags in colony weight gain, with the maximum difference being a doubling of exposed hive weights compared with more than a six fold increase in control colonies in 5 weeks (Greenberg et al., 1981a). Foraging rates were decreased by as much as half in exposed colonies in this study (Greenberg et al., 1981a). Honeybees also had an increased tendency to sting under high voltage transmission lines (Wellenstein, 1973). ELF EM fields might similarly affect megachilid bee activity by disorienting or agitating the bees so that the duration of leaf- and pollen-foraging trips is altered. Interference with magnetoreception might play a role in disorientation. Changes in electric potential of the bees, or of the plants on which they forage (Erickson, 1975; Erickson and Buchmann, 1983), or changes in the electric potential of antennal chemosensilla that detect plant odors (Erickson, 1982) might also affect the bees' foraging rate.

Leaf-foraging trips for *M. inermis* are easy to recognize behaviors, usually lasting less than a minute in duration. Many of these trips are taken in succession, so within and between bee variability can be analyzed, and a potentially large sample of leaf collecting trips can be timed. In the 1986 Annual Report we demonstrated that the collection of LO leaves was the most consistent behavior of the leaf-cutting bees under study. We argued that this is probably because it is adaptive to close the cell as quickly as possible after the egg is laid to avoid parasitism. Thus, our analysis focuses on LO trip durations.

Hypotheses Involving Emergence:

Hypothesis 6: Overwintering mortality of megachilid bees is unchanged by exposure to ELF EM fields.

Rationale

Overwintering mortality of honeybee colonies under high voltage transmission lines increased from 29% when hives were shielded to 71% when they were fully exposed to electrical fields. (Greenberg et al., 1981b). We would like to test for a similar effect in megachilid bees. To do this requires comparing control and experimental sites in the proportion of cells that suffer mortality during the prepupal (overwintering) stage, relative to the number of cells that survive to the prepupal stage or beyond (pupa and adult) (see results section for further discussion).

III METHODS AND TYPES OF DATA COLLECTED

Nest architecture and nest orientation are obtained by placing trap nests in the environment, and allowing bees to construct nests in their choice of traps during the summer. The following spring, various parameters of their nest architecture are measured. Bee and parasite emergence and larval and pupal mortality are also recorded in the spring. Nest activity data are gathered during the summer season while the bees are constructing their nests.

The methods discussed below will compare, where appropriate, changes in protocol over the years, especially pre- and post-1987. Where no such comparisons have been made, no significant changes in protocol have been made.

Trap Nesting Methodology

Bees are provided with fresh trap nests each year. Trap nests consist of elongate white pine pieces 19x19x153 mm. Most of these nests were drilled lengthwise to a depth of 142mm. Exceptions were the largest diameter nests pre-1987, and half of the 1987 large diameter nests. These nests were drilled to only 107mm.

Prior to 1987, drill bits with seven different diameters were used to create trap nests (Table 1). The maximum diameter was limited by the dimensions of the trap nest, and by availability of long drill bits.

In 1987 only the 5.5mm bit and the 11.0mm bit were used because these diameters were accepted most often in 1985 by the two *Megachile* species under study (see 1986 annual report). In 1988-1991 small nests were made with both 5.5 and 6.0mm drill bits because analysis of 1986 nests indicated that the 6.0 mm diameters were common, and because it was feared that 5.5 mm diameters would skew the sex ratio in favor of male offspring and thus bias the cells towards shorter lengths. Bore diameter has been shown to influence sex ratio for other trap nesting species (Stephen and Osgood, 1965; Krombein, 1967; Cowan, 1981; Tepedino and Torchio, 1989).

Prior to 1987, twelve nests, two of each bore diameter, were bound together with plastic strapping into a "block", so that one of each bore diameter faced each direction, and no two bore entrances were adjoining (Fig. 2a). Starting in 1987, two 11mm bores and four 5.5mm bores were arranged randomly in each direction (Fig. 2b). In 1988-1992, three of the small nests were 5.5mm and one was 6.0mm in each direction. We did not realize that the 1987 random arrangement of nest entrances differed from the 1983-86 pattern of no adjoining entrances

until blocks for 1987 had already been prepared. We have observed no obvious changes in bee behavior at the hutches as a result of this change in nest entrance arrangement, although we have made no systematic effort to compare the two arrangements.

"Hutches" consisting of a wooden frame with four shelves and a roof were used to hold the blocks of trap nests (Fig. 3). Four blocks of nests were placed randomly on each shelf, making a total of 192 nests present at any one time. The hutch was open on both sides, so half of the nests opened in each direction. The shelves were roughly 0.1, 0.4, 0.8, and 1.1 meters from the ground.

Four study sites were selected by 1984 for placement of hutches (Fig. 4). Two are experimental sites along the ELF antenna: Ford 1 and Ford 2 (F1 and F2), and two are control sites: Camp 5 and County Line (C5 and CL). The study sites are described in the section titled "Description of Sites", p. 20. Further information can be found in the 1985 annual report. Three sets of two hutches, making a total of six hutches, were placed at each of the four study sites. In each set of two hutches, one hutch was oriented in a north-south direction so that its nests open to the east or west, and one hutch was oriented in an east-west direction so that its nests open to the north or south. The two hutches in each set were placed near each other in edge habitats between open areas where there are abundant flowering plants, and woods where natural nest sites are available. In 1983, only the F1 site had been chosen for study in the spring. The CL and F2 sites were added in mid-season. Generally, only one or two sets of hutches were in place that year.

When a nest was occupied by a megachilid bee, it was given a number that included site (C5, CL, F1, or F2), hutch direction (NS or EW), nest entrance orientation (E, W, N, or S) and shelf height (1-4, top to bottom). This number was written on the side of the nest. Position on the shelf and in the block of nests was not recorded. Starting in 1987, a computer data base was created to help us manage nest numbers and progress of the nesting bees.

Once a nest in progress was identified, the depth of empty tunnel space was recorded daily (pre-1987) or every 2-7 days (1987-92). This information, coupled with nest architecture measurements taken the following spring, allowed us to estimate which cell the bee was constructing on the day the nest was first located. Assuming that the bee takes approximately one day to complete a cell, we estimated the dates on which the nest was begun and finished. Nests were classified as "early season" if they were begun on or before the date on which half of the nests of that species (pooled over all sites) were begun

during that year. Nests begun on later dates were classified as "late season" nests. When the nest was completed, it was removed from the block, and replaced with an empty nest of the same bore size.

Each completed nest was stored in a large centrifuge tube with cloth covering the opening. Tubes were placed in wooden overwintering boxes built to fit the hutch shelves. Prior to 1987, completed nests were brought to Channing to overwinter, in order to avoid vandalism and marauding animals. However, starting in 1987, nests were left in overwintering boxes at the site where they were constructed. Overwintering boxes were not left on hutch shelves as in the past, but rather were elevated about a foot off of the ground and camouflaged with branches, bark, and leaves in order to avoid vandalism. Fortunately, overwintering boxes have not been vandalized at any of the sites, although hutches have been damaged and have disappeared during the winter.

Beginning with nests constructed in 1990 and continuing in 1991, a manipulative experiment was initiated to compare overwintering mortality of nests constructed at one site but overwintered either at an experimental or a control site. The results of this experiment cannot determine unambiguously whether ELF EM fields affect overwintering mortality, but the experiment may offer further evidence in conjunction with broader comparisons between all sites and years. For the manipulative experiment, each year one third of the nests constructed at the F2 experimental site were moved to the C5 control site in mid-September for overwintering. The nests that were moved were chosen to represent hutches and dates of nest initiation in the same proportions as the nests that remained at the F2 site. The number of F2 nests overwintering at C5 approximately equaled the number of C5 nests overwintering at C5. Nests from both sites were placed in overwintering boxes in the same directions as they were constructed, but C5 and F2 nests were mixed and positioned randomly with respect to bottom vs. top, right vs. left side of the overwintering boxes. The reciprocal experiment, overwintering C5 nests at F2, could not be conducted in 1990 or 1991 because there were insufficient C5 nests. Nest numbers were considerably reduced at all sites in 1992, so there were insufficient nests to continue the experiment.

Nest Architecture Measurements

Nests constructed by *M. relativa* during 1983 were measured in the spring of 1984 prior to emergence. Nests constructed by *M. relativa* during 1985 were measured after bee emergence, in November and December, 1986. Nests constructed during 1985 by *M. inermis* were measured after emergence in August, 1987. Most

1986 *M. relativa* nests were measured before emergence in 1987, so that we would know with certainty the species and sex of the occupant of each cell. The 1986 *M. inermis* began to emerge in spring 1987 before we began measuring their nests, so most *M. inermis* nests were measured after bee emergence. The 1987-91 nests were measured sufficiently early in May of 1988 - 1992 that we were able to complete nest measurements of both species before they emerged in June and July.

Measurements for 1986-1990 nests were made at our Crystal Falls lab. However, we learned in 1989 that 60hz EM fields are relatively high in Crystal Falls due to the presence of numerous power lines. In the laboratory, electric lights and wiring in the walls also create relatively high EM fields (ELF Communications System Ecological Monitoring Program: Electromagnetic Field Measurements and Engineering Support - 1990). Therefore, beginning in 1989 unopened nests and rearing tubes were kept at a holding site constructed by the ELF Small Mammal and Bird Project in woods 5 miles south of Crystal Falls. Nests were brought to the Crystal Falls lab only briefly for measurement. There they spent up to 6 hours outside the house where 60 hz fields were low, and no more than 2 hours in the lab for measurements. In addition, in 1990 and 1991, measurements were made in wire mesh Faraday cages constructed by IITRI to minimize exposure of developing bees to electric fields (Fig. 5). Just before and just after measurements, nests and cells were stored in another Faraday cage on the front porch of the Crystal Falls Lab (Fig. 6). In 1992 we moved our research to a smaller house in Crystal Falls because fewer assistants were required. Unfortunately, EM fields were considerably higher in this new house, so all nest architecture measurements were made at the holding site.

After recording nest number and bore diameter, nests were split open lengthwise with a chisel. Total bore depth, non-reproductive spaces (basal space, vestibular spaces, associated caps, nest plugs, and indentation) were measured with the cells intact. Each cell was then removed and measured from the base of the cell to the position of the outermost leaf in the cell cap (Fig. 7). Cell lengths measured after emergence are likely to be somewhat more variable than cell lengths measured before cell emergence, because emergence damages the cell cap. Thus it is sometimes difficult to determine where the edge of the cell cap starts.

The nest number that is written on each nest includes information on the site where the nest was created, so nest architecture measurements of pre-1988 nests were not blind to site. We doubt that knowledge of the nest site affected our measurements. However, in response to reviewer concern, our measurements of the 1988 - 1991 nests were made blind to site. Before nest measurements were made, students who did not

measure nests spent a day crossing out nest numbers and replacing them with a random number independent of site. A data base not available to the nest measurers recorded the original nest number, and the random code number assigned to it. Nests were then measured without knowing at which site they were constructed. After all measurements were complete, the random number was associated with its original nest number, including site.

Since more than one person measures nests, we attempt to divide the nests equally by site and date of nest initiation among all measurers. Thus individual biases in measurement are distributed evenly between sites and dates. In addition, in 1987 thirty-nine *M. relativa* cells were re-measured to determine within- and between-individual measurement error. Twenty cells were measured three times by each of the four individuals measuring nests. An additional 19 cells could only be measured 1 or 2 times by each measurer, because they were damaged by the multiple measurements. Similar experiments were repeated in 1990 and 1991, with 3 measurers and 39 *M. inermis* cells from nests constructed in 1989 and 1990. Each cell was measured 3 times.

Emergence Data

Nests created in 1985 were checked daily in the spring of 1986 for bees that had emerged from the nest and were in the centrifuge tubes. In subsequent years, after taking nest measurement in the spring, cells from which nothing had yet emerged were placed in individual plastic culture tubes or 2 oz. transparent plastic "Solo" rearing dishes, and labeled with nest and cell identification numbers. In 1987 and 1988 tubes were kept in the Crystal Falls Laboratory at room temperature (approx. 68°F) until emergence. Beginning in 1989, cells in rearing dishes and culture tubes were returned to the holding site in woods 5 miles south of Crystal Falls after nest measurements were complete. Cells were checked daily for emergence. In all years, date of emergence, species, and sex of offspring were recorded. Live weights were also obtained for most bees from 1988 - 1991 nests (see below).

Some bees were saved for dry weight measurements (see below) and identification. Bees were identified by G. Dahlem, V. Scott, and K. Strickler based on Mitchell (1962), and by comparison with reference specimens provided by T. Griswold, ARS Bee Laboratory, Utah State University, Logan Utah.

The remaining adult bees were released at the sites where their nest had been constructed the previous summer. The Faraday cages mentioned above were intended to insure that released bees were not affected by 60 hz electric fields when

nest architecture measurements were taken. Effects of 60 hz fields might be mistaken for (or might mask) effects of the ELF antenna's 76 hz fields, and affected bees might alter the genetic makeup of natural populations. Parasites were collected and not released. Also, F2 nests that had overwintered at C5 were not released at the research sites.

Cells that showed no signs of emergence were opened in August (1986-91 nests), or when the nest was measured (1985 nests). Contents were recorded to indicate at what stage mortality had occurred.

Offspring Weights. Weight may be a more appropriate measure of parental investment per offspring than is cell length. Weight depends directly on the amount of provisions in the cell (see introduction, p. 2), and provisioning the cell typically takes much more time than constructing a cell. A larger cell with more leaves may have less provisions and thus yield a smaller offspring than a smaller cell with more provisions, which cost more in time and energy to gather than the leaves.

Some authors (eg., Cane, 1987) and one of our reviewers have suggested that measurement of hard body parts is a better indicator of body size than is weight, although the two are correlated (Cane, 1987). Field collected bees, which vary in age and foraging status, may be especially variable in weight. Since bees in this study were collected within hours of emergence without being released, their crops were empty. Thus much of the variability in weights that would be expected from a sample of field collected bees was eliminated. This year we measured a sample of bees to see how closely correlated weight is with measurements of body parts. We measured head width, intertegular width on the thorax, and tibia length (Fig. 8) of 30 individuals of each sex for each species.

For 1988-1991 nests, live weights of all female and most male *Megachile* were measured with a Sartorius H51 balance (accurate to 0.1mg) on the day that they emerged. When emergence took place at the holding site, live weights of most bees were taken in the field. In addition, two or three bees from each 1986 - 1991 *M. relativa* nest were saved for dry weight measurements and for confirmation of species identification. In 1988-92, *M. inermis* individuals from 1987-91 nests were similarly saved for dry weight measurements. Dry weights were obtained by drying bees in a desiccator over P_2O_5 to constant weight. Constant weight was defined as two weights taken 48 hours apart that were within 0.5mg of each other. The lower of these weights was used in analyses.

Offspring sex, expected sex of a cell, and sex ratio. Previous analyses indicate that the offspring's sex contributes significantly to variance in cell length and leaves per cell (1989 and 1990 Annual Reports). However, the sex of the offspring was known only for a small proportion of the cells, since many offspring die in the larval and prepupal stages, which can't be sexed. Furthermore, parasites emerge from some cells rather than *M. inermis* or *M. relativa*. In an attempt to increase our sample size, we create a new variable in our data set that indicates the expected sex of a cell. We can predict the expected sex for many of the cells that did not have a bee emerge. Emergence data for 1987-1991 nests indicate that when a nest contains females, they are almost always in inner cells relative to cells containing males. Very few males have ever emerged from cells that were deeper than a female cell: only 4 *M. relativa* and 7 *M. inermis* in 1987, one *M. relativa* and 9 *M. inermis* in 1990, and one *M. relativa* and 3 *M. inermis* in 1991. Therefore, cells of unknown sex deeper in the nest than a cell with a female offspring can be assumed to be female. Conversely, cells with unknown sex that follow a male cell can be assumed to be males. Similar deductions have been used in analysis of sex ratio in other studies of trapnesting bees and wasps (Cowan, 1981; Sugiura and Maeta 1989).

The expected sex of a cell is the predicted sex of the cell when sex can be deduced, or the actual sex when sex is known. In statistical analyses where female and male cells are treated separately, expected sex increases the number of cells that can be included in the analysis by 2.6 fold for 1985 *M. relativa*, by 2.4 fold for 1985 *M. inermis*, and by 1.2-1.8 fold for both species in subsequent years.

Expected sex of a cell is a useful variable in analyses of cell length and leaves per cell. However, it is not a good variable to use in estimates of sex ratio of the population. This is because expected sex cannot be deduced in nests that have only a single dead cell, or in nests that have no emergence in the innermost cell and only males in subsequent cells. Since the innermost cell has the highest proportion of female offspring, using expected sex of a cell to estimate sex ratio will bias the sex ratio toward males. Instead, we use the ratio of male to female adult and pupal offspring that could be sexed with certainty.

Leaf Counts

The number of elongate leaves that were used to construct a cell was determined for 1985-1990 *M. inermis* cells and 1986-1990 *M. relativa* cells that were still in good condition once emergence was complete. Leaves lining *M. inermis* cells overlapped, but were easy to tease apart and count. Leaves

lining *M. relativa* cells were smaller, and were fastened together so that a microscope was often needed to determine where one leaf ended and the next began. When in doubt, leaf counts for *M. relativa* cells were not recorded.

Data Entry for Nest Architecture, Emergence, and Leaf Count Data

Nest architecture measurements, emergence records, and leaf counts were recorded manually in the spring and summer on data sheets for each nest. During the past year we have been moving our data base management and statistical processing from the Entomology Department's old and outdated VAX 11/730 computer to a new 486 33mhz PC computer. Last year the VAX machine crashed several times while we were analyzing data, causing great inconvenience for us. Now, nest architecture data are typed into an R-Base database management file, taking the place of INGRES on the VAX. Relevant subsets of the data are transferred from R-Base to SAS data files for statistical analysis with PC SAS. This year we have continued to use SAS GRAPH on the VAX computer for creating some of our figures because we do not yet have that program for the PC.

Nest Activity

One or more observers have gathered data on behavior of individual bees at the nest every year between 1983 and 1991. No new data were gathered this year. We are updating data from 1983-1986 to make them compatible with data collected since 1987. We have not yet analyzed these data however. What follows is a repetition of our explanation of nest activity methods from last year's annual report.

In the 1986 Annual Report, we decided to focus on the collection of round pieces of leaf (LO trips) used in capping a cell. Analysis (1986 Annual Report, p. 20-21) suggested that this was the most consistent of the three main behaviors in nest construction (collection of pollen, collection of elongate leaves for cell lining, and collection of round leaves for cell caps). LO trips probably involve fewer extraneous behaviors such as sunning or taking nectar than do pollen or elongate leaf collecting trips. Thus residuals for the transformed duration of LO trips could be normalized for statistical analysis. Consistency in LO trip durations probably results from the necessity to cap the cell rapidly to avoid parasitism after laying an egg.

Prior to 1987 each observer watched a single bee for several days in succession, until the nest was complete. This protocol generated a great deal of information on the variabil-

ity in behavior within a bee, but less information on between-bee variability. In 1987 - 1991 field seasons we maximized the number of bees timed per day, rather than timing one bee for long periods of time. Observers became adept at locating a bee that was about to lay her egg, and were able to focus on timing the first few LO trips that the bee made after laying her egg. Generally, we tried to time 5 such trips in succession before searching for another bee that was about to collect LO leaves. Occasionally the bee would complete a cap in fewer than 5 timings. The observer sometimes would time more than 5 LO trips if no other bees were active. Number of trips timed for a bee on a given day ranged between 1 and 18. In 1987 we did not keep track of the number of LO trips that the bee had made since an egg was laid before we began timing. Our 1987 analysis suggested that this "trip rank" number is important (1987 Annual Report), because LO trips tend to increase in duration with each successive trip after egg laying. Thus, during the 1988-1991 field seasons we attempted to record this number when timings were made. Only LO durations for which the trip rank order was known are used in the current analysis. Furthermore, no more than the first three trips are included in statistical analysis, because this minimizes the variability in the duration of a given bee's cell capping trips.

During the 1987 - 1991 field seasons, four observers were rotated between sites every 3 to 4 days, so that biases between observers would be distributed evenly between sites and dates. On a given day, two observers visited a control site and two an experimental site.

Prior to 1987, the duration of LO trips was determined by using a watch to record the hour, minute, and second that the bee left the nest and returned to the nest. Since 1987, we have used portable Tandy 102 computers that are programmed as event recorders. When the program is activated, the observer is prompted for information on the nest number and site, and some weather data (see below). The program automatically numbers the observed activities in sequence. Hitting the space bar records the time to the nearest second at which the bee leaves the nest or returns to the nest. A single letter code is used to indicate what cargo (e.g., LOs), if any, the bee brings back to her nest. An editing feature allows the observer to correct errors made during the timings, or to delete times that result from hitting the space bar inappropriately. These data were down-loaded to a Zenith personal computer at our field headquarters, and later transferred to an INGRES data base file on the VAX computer in the Department of Entomology at MSU. Duration of each trip was calculated in INGRES by subtracting the time when the bee left the nest from the time when the bee returned.

Weather Data

Because behavior of insects is often affected by such environmental factors as temperature and wind speed, foraging trip durations might be correlated with weather conditions. Some weather data were recorded in the event recorders as each bee was timed during the 1987-1991 field seasons. This included sun conditions (sunny, partly cloudy, cloudy, rain), temperature in the shade on the same shelf as the bee's nest, shading of the block in which the bee's nest was found, relative humidity calculated with a sling psychrometer, average wind speed and speed of wind gusts measured with a Dwyer Portable Wind Meter (hand held). These weather data were downloaded from the Tandy computers to the Zenith, and then to an INGRES file on the VAX computer (as described above for LO durations). We are currently in the process of transferring weather data to an R-Base file on the 486 PC, and from there to a SAS data set.

Data on long-term trends in temperature and precipitation were also obtained from the ELF Herbaceous Plant Cover and Tree Studies project, based at MTU. Dr. Hal Liechty of the MTU project kindly provided us with an asci file of daily summaries of average, 3 hr. minimum, and 3 hr. maximum air temperatures, and total daily precipitation. Ambient monitoring of air temperature and precipitation (among other variables not of interest to us) takes place at MTU's Red Pine Plantation sites: a treatment site under the ELF antenna, 10 miles North of our F1 site; and a control site 9 miles south of Crystal Falls. Despite the distance between the MTU sites and the sites that we are using in the Native Bee ELF project, major climatic trends and differences between years in temperature and precipitation are representative for the region. Climatic trends correlate with floral resources and thus with bee population size, cells per nest, offspring weight, and percent mortality. For further information on the MTU ambient monitoring system, see Appendix B of the 1985 Herbaceous Plant Growth and Tree Studies Project Annual Report.

Description of sites

Figure 4 shows the location of the study sites relative to the ELF antenna. Three sites are located on Copper County State Forest Property in Dickinson Co. in the Upper Peninsula of Michigan. A fourth site (C5) is located in Iron Co. on property leased by the Michigan Department of Natural Resources to Champion Paper Company. Permission to use these sites is gratefully acknowledged.

The C5 site is located 6.7 km south of Route 69 and about 0.8 km west of Camp 5 road in Iron County, Michigan (Township

42N. Range 31W, Section 14). The area has recently been logged, and nearby forests continue to be logged within a km. of our hutches. An abandoned railroad bed runs N-S through the site. Camp 5 creek runs through the site, creating a cut-over swamp and flood plain. Two hutches are located at the south edge of this flood plain, and two hutches are located in an open depression next to the abandoned railroad bed. Until mid July 1990 the last two hutches were at the north edge of the flood plain, north of C5 creek. This site was not close to *Cirsium palustre* populations, and attracted few *M. inermis*.

In spring, 1990 a beaver made a dam across C5 creek, making access to the north hutches impossible by crossing the creek next to the railroad right-of-way. For several months we walked around the edge of the flood plain to reach the north hutches. However, as the water behind the dam increased, the flood plain turned into a shallow lake. On July 25, when water was within 10 feet of the north hutches, we moved them to the south side of C5 creek. The hutches were relocated to an elevated site about 20 feet west of the railroad right-of-way, near a large patch of *Cirsium palustre*. The bee population that uses nests at these hutches should be the same as in the original location. However, being closer to flower populations, more bees are nesting at the new location.

Nearby woods consist primarily of *Populus tremuloides*, with occasional *Larix decidua*, *Picea glauca*, and *Prunus serotina*. Shrubs in the vicinity include *Alnus rugosa*, *Vaccinium* sp., *Salix* sp., *Spirea alba*, and *Rubus allegheniensis*. Herbaceous plants include *Cirsium palustre*, *Fragaria virginiana*, *Hieracium* spp., *Trifolium* spp., and *Solidago* spp.

The CL site is located about 1.7 km north of Route 69 on the east side of County Line Road, in Dickinson Co., (Township 43N, Range 30W, Section 19). Logging continues within a km or so of the hutches. This site has very sandy soil and is the driest of our sites. Hutches are located at the edge of clearings in *Populus tremuloides* woods, with occasional *Acer saccharum*, *Betula papyrifera*, *Abies balsamea*, and *Pinus resinosa*. Two hutches are adjacent to a patch of trees north of a logging road through the sandy clearing. Two are east, and two west of a marshy, low lying area south of the logging road. *Hieracium aurantiacum* carpets the ground at this site in June, if rain has been sufficient. Bracken fern is common near the east hutches which are in a shadier location than the others. Other flowering plants that are common in the area include *Cornus canadensis*, *Campanula rotundifolia*, *Fragaria virginiana*, *Rubus* spp., *Solidago* spp., *Vaccinium* spp., and *Prunus pensylvanica*. Small patches of *Cirsium palustre* grow in the marshy area south of the logging road. *Epilobium angustifolium* was abundant at this site in 1983, but decreased rapidly

thereafter. Only a couple of stems were present in 1987, and none in subsequent years.

Low numbers of *M. inermis* nests at the CL site, especially in 1986, prompted us to transplant about 90 *Cirsium* spp. plants (a common pollen source at other sites) to the CL site in April, 1987 and 50 plants in June 1989 to try to increase the numbers of *M. inermis* that nested there.

The F1 site is located south of Turner Road, and north of the Ford river, 20 km east of Channing. (Township 43N, Range 29W, Section 14). The hutches are located at the edge of a flood plain, bordered on the north by a Red Pine plantation, and the south by vegetation along the river consisting of *Populus balsamifera*, *Populus tremuloides*, *Fraxinus nigra*, and *Alnus rugosa*. A corridor has been cut through the pine plantation for the ELF antenna, which runs NE-SW through the site. Two hutches are east of the antenna, at the north edge of the flood plain. Two are a similar distance west of the antenna. Two are in a shady clearing further west of the antenna at the northwest edge of the flood plain. Flowering plants near the hutches include several species of *Cirsium*, especially *C. palustre* and *C. arvense*, *Urtica dioica*, *Solidago* spp., *Hieracium aurantiacum*, *Hypericum perforatum*, *Aster* spp., *Rubus* spp., *Humulus lupulus*, *Linaria vulgaris*, and *Vaccinium* spp.

The F2 site is located about 0.8 km south of the Ford River and the F1 site, along the clear cut for the ELF antenna. The soil is sandy. Three of the hutches are located on top of a hill at the edge of the clear cut west of the antenna, and along an old logging/hunting trail running west from the antenna. Three hutches are located in a valley east of the antenna. Nearby woods consist of *Populus tremuloides*, with occasional *Picea glauca*, and *Pinus resinosa*. *Centaurea maculosa* has increased since 1983 until it is now the most abundant flowering plant on the hill. Also abundant are *Cirsium palustre*, *Fragaria virginiana*, *Hieracium aurantiacum*, *Coronilla varia*, *Prunus virginiana*, *Rubus idaeus*, *Solidago* spp., and *Trifolium* spp.

ELF Antenna Operations

In interpreting results of this project it is important to know the pattern of antennal operations in past years (Fig. 9, 10). The Michigan Transmission Facility (MTF) began testing at 10% power (15 amperes) periodically during the summer (March - October) of 1986, and with increasing regularity from May - November, 1987 and January - July, 1988. Starting July 6, 1988 and lasting until May, 1989, testing continued at 50% power (75 amperes).

In June 1989, the ELF antenna began testing periodically on full power (150 amperes). Continuous full power operation began in October, 1990.

Cumulative potential magnetic field exposure of the bees is plotted in Figs. 9 and 10, based on measurements provided by IITRI (Technical report, ELF Communications System Ecological Monitoring Program: Electromagnetic Field Measurements and Engineering Support). Unlike electric fields, magnetic fields are not blocked by trees, hutches, or trap nests. Thus magnetic fields are more likely than electric fields to affect the bees. Gauss-Hours of magnetic field exposure of foraging bees during June, July and August are plotted in Fig. 9. A bee sitting directly under the antenna for the entire month would experience the maximum exposure plotted. A bee sitting on the hutch farthest from the antenna at the F2 site would experience the minimum exposure plotted (solid bars). Most bees at the experimental sites would experience intermediate magnetic field exposures while foraging. Figure 10 plots Gauss-Hours of magnetic field exposure of bee prepupae in overwintering boxes between Sept. and April at the two experimental sites. Exposure of the prepupae at the F2 site was approximately twice that of the F1 site in 1989 and 1990. Insufficient data for 1991 nests were available at the time of this writing, but magnetic field exposures were probably very similar to 1989 and 1990 exposures.

In this year's analyses, 1989 - 1991 are considered full power years. Exposures during the intermediate years of 1986-1988 were considerably lower than in subsequent years, and analyses did not show any effect of experimental and control areas on nest architecture and activity. Therefore, we analyze 1986-1988 data as pre-treatment years.

Statistical Methods

The General Linear Models (GLM) procedure on SAS for PC (Version 6.04) was used to analyze sources of variability in cell lengths (both species), nest plug length (*M. inermis*), leaves per cell (*M. inermis*) and LO trip durations (*M. inermis*). Because cell lengths or leaves per cell within a nest, and/or LO durations within a cell capping bout, are autocorrelated, we calculate a mean cell length or leaf number for each nest, or a mean LO duration for the first three LOs in a cell capping bout. GLM analysis was accomplished on these means. In this model, the error variance consists largely of between nest variability.

In GLM analyses, means of LO duration per cell capping bout were weighted by the number of trip ranks (1-3) that were used to calculate the mean. However, means of cell lengths and leaves per cell were not weighted by number of cells per nest. Rather, cells per nest was a covariate in the models of cell length and leaves per cell.

Incomplete cells (without a cell cap) were not included in calculations of mean cell length for a nest. Not all cells could be measured in some nests, because some of the cells were destroyed by emerging bees. This may have biased the mean cell length of the nest, if most of the unmeasurable cells were inner cells or outer cells. No attempt was made to adjust for such possible biases.

Table 2 summarizes the GLM model that was used to analyze cell lengths, nest plug lengths, leaves per cell, and LO durations. Fixed main effects included experimental vs. control areas (referred to as "Exp"), and Antenna operations (Off=1983-1988 or On=1989-1991). Random nested effects included sites (Site[Exp]), observers or measurers nested in year, and years nested in antenna operations. Where appropriate, other fixed class variables included to explain the variability in the dependent variables were the "expected sex" of a cell, complete vs. incomplete nests, and early vs. late season nests. Number of cells per nest and nest diameter were covariates in the analysis of cell lengths, nest plug lengths, and leaves per cell. Date of the trip was a covariate in the analysis of LO trip durations. Time was tested as a second order covariate in this analysis. Significance would indicate that LO durations are faster (or slower) during the middle of the day, as might be the case if LO durations are correlated with temperature. Type IV mean squares were calculated in all GLM analyses. This model is invariant to the ordering of effects in the model.

The mean square (MS) of Site[Exp] was included in the error term for testing the significance of Exp. This insures

that differences between experimental and control areas are significant only if they are greater than any differences between the sites within the areas. The MS of Measurer [Year * Antenna] was included in the error term for testing Year[Antenna], and both Year[Antenna] and Measurer [Year * Antenna] MS were included in the error term for testing the antenna main effect. This insures that differences between years are greater than the differences between measurers who took data in any given year, and that differences between "on" and "off" antenna years are greater than the differences between years within those time periods. The GLM procedure calculated appropriate error terms and degrees of freedom for these mixed model analyses using the Random/test statement.

The most important effect in the GLM model is the interaction term Exp*Antenna. If significant, this term indicates that the magnitude of the difference between treatment and control areas is different during "on" antenna years than during "off" antenna years. If the Exp main effect is significant but not the Exp*Antenna interaction, then we know that there are intrinsic differences between experimental and control areas that have nothing to do with the antenna. If the antenna main effect is significant but not the Exp*Antenna interaction, then we know that there are differences between "off" years and "on" years that have affected both experimental and control areas equally, as would be the case for climatic changes between years. If the antenna is having an effect, treatment areas should change more between "on" and "off" years than do control areas. If control areas change more than treatment areas between "off" and "on" years, there is some ambiguity, and we must consider whether the treatment areas should have changed but did not because of ELF EM fields, or whether there is simply more variability between control areas than between treatment areas. Furthermore, because sample sizes are very large, we must also consider whether a marginally significant effect has biological meaning.

The interaction term Exp*Antenna was tested with the model error term. In previous Annual Reports we mistakenly used the Site[Exp] mean square as the error term. Examination of Appendix A in Zar (1984, pp. 470-476), consultation with Dr. D. Gilliland of MSU's Statistics Department, and results of the Random/Test statement in the SAS GLM procedure convince us that our current approach is more appropriate than the previous analyses.

A Shapiro-Wilk statistic for $N < 51$ and a Kolmogorov D statistic for $N \geq 51$ in the Univariate procedure of SAS were used to test for normality of residuals in GLM models. The data are tested against a normal distribution with mean and variance equal to the sample mean and variance. The significance level used in these tests was $\alpha = 0.05$. Ln or lnln

transformations of the data were sometimes required to meet the assumption of normality of residuals. When used, such transformations are discussed in the Results section. In some cases where residuals were significantly different from normal, a plot of the residuals revealed that a few outliers or a slight skewness of the data were responsible. In these cases the GLM results are likely to be robust, so they are reported. An alternative non-parametric test was tried on nest plug lengths (Zar, 1984 p. 250-251, 221). Lengths were ranked, and the usual GLM model was calculated on the ranks. An H statistic was calculated by dividing the sums of squares from the GLM model by $N(N + 1)/12$, where N is the total sample size. This statistic is closely approximated by χ^2 with degrees of freedom appropriate to the source of variation being tested.

Minimum detectible differences between experimental and control areas (Exp) were estimated with a modification of Cochran and Cox's (1975) formula (Zar, 1984 p.135, 137, 260). Sample size used in this formula was the harmonic mean of the treatment and control area sample sizes (Zar 1984, p. 137) based on numbers actually collected each year for the control and experimental areas. The value of population variance s^2 , used in calculating minimum detectable differences was the denominator mean square calculated by the GLM procedure for Exp (Zar, 1984, p. 260). Values of α and the power of the test ($1 - \beta$) were 0.05 and 0.9 unless otherwise stated. When the Exp*Antenna interaction was significant, the actual power of the test was calculated using the procedure described by Zar (1984 p. 227).

A two-way classification model II ANOVA was used to analyze within- and between-measurer components of cell length variability (Sokal and Rohlf, 1969, p.313). In this analysis, measurer and cells measured were random effects. The error mean squares gives within-measurer variability.

The Categorical Data Modeling (CATMOD) procedure on SAS was used to compare distributions of cells per nest, the proportion of fly parasites vs. other emergences, the proportion of incomplete vs complete nests, and the proportion of males vs. females emerging. This statistical program fits linear models to functions of response frequencies for discrete data; i.e., it is an extension of the GLM procedure for continuous data. The program uses a Wald statistic (which approximates a chi-square distribution for large sample sizes) to test hypotheses about linear combinations of the parameters in the model. As with the GLM tests previously described, we tested for significance of experimental vs. control areas (Exp), Sites nested in Exp areas (Site [Exp]), Off vs. On years (Antenna), Year[Antenna], and the interaction between Exp and Antenna (Exp * Antenna). However, a mixed model analysis as

was used in GLM analysis is not available with the CATMOD procedure in SAS. Early vs late season were also included in some analyses. The level of significance of all tests was $\alpha = 0.05$. Because of small sample sizes for some site-year categories, we use maximum-likelihood estimates in testing our models.

Proportion of nests oriented in a N-S vs. E-W direction was tested in a log-likelihood ratio contingency table analysis (Zar 1984, p. 67-68) to determine if the pattern of directions of nests was the same for all years at a given hutch set. If consistency was found between years, then data for a hutch set were pooled over years, and tested against other hutch sets at a given site. If the ELF antenna was affecting choice of nest direction, then the contingency tests should be significant at some or all of the hutch sets at experimental sites, but not at the control sites. In addition, a change in nest orientations should occur some time between 1988 and 1990. Prior to the change, nest orientation should have been consistent over pre-operational years. Similarly, any changes that occur as a result of ELF EM fields are expected to continue during subsequent operational years.

Mortality in the overwintering prepupal stage was tested with a CATMOD analysis comparing prepupal mortality with survival to at least the pupal stage. The model used is the same as that described above for cells per nest. Calculation of proportion of prepupal mortality was complex, and will be explained further in the results section.

In past annual reports, we transformed proportion of mortality in the prepupal stage using a Freeman and Tukey arcsine transformation, which is particularly useful for small proportions (Zar 1984, p. 240):

$$P_{ft} = \frac{1}{2} \left(\arcsine \sqrt{\frac{X}{(n+1)}} + \arcsine \sqrt{\frac{(X+1)}{(n+1)}} \right)$$

Resulting values were analyzed with the GLM procedure in SAS. This year we used the arcsine transformed proportions in a Tukey test to determine which sites and years were significantly different.

CATMOD analysis was used to compare prepupal mortality for 1990 nests moved from F2 to C5 to overwinter. Nest construction site, overwintering site, and nest direction were tested in the model.



IV NEST ARCHITECTURE RESULTS

Climate, Floral Resources, and Bee Abundance

Table 3 & 4 and Figs. 11 and 12 summarize the number of nests of the two species for which we have data on cell lengths, and an estimate of the number of complete nests created in 1992. Some 1985 *M. inermis* nests were not included in our measurements because Dr. Fischer, who initiated this research project, used them in experiments on diapause. Data for 1983 are available only for *M. relativa*. Nests were monitored for the entire season only at the F1 site, and only at two hutches at this site. Some information is also available for late season nests at CL and F2 in 1983. Unfortunately, data for 1984 nests were either not taken, or were unreliable due to personnel problems at the time. No further nest measurements will be made for this project. However, 1992 nests will provide a final year of data on cells per nest.

Number of nests were low for both species at all sites in 1992. Between 1983 and 1992 *M. relativa* has produced similar numbers of nests at all sites (17-128), with no consistent differences between control and treatment sites (Fig. 11). In contrast, *M. inermis* produced a consistently lower number of nests at the control sites, especially CL, than at the experimental sites (Fig. 12). Furthermore, *M. inermis* nest numbers at all sites were lower in 1986, 1988 (except for F2), and 1992 than in other years. We believe that these reductions in *M. inermis* populations were caused by a reduction in floral resources due to low rainfall, especially early in the season. Fig. 13 plots cumulative precipitation for 1985- 1992. The first nests of *M. relativa* and *M. inermis* are indicated on the plots, along with first bloom (when known) of two important pollen plants for the bees: *Hieracium aurantiacum*, and *Cirsium palustre*. In 1986, 1988, and 1992 bee nesting and plant flowering began when less than 4 inches of rain had accumulated, whereas in 1985, 1987, and 1989 - 1991 the same events began after 4-5 inches of rain had accumulated. Although no quantitative measures of numbers of flowers in bloom were made, we did note that *H. aurantiacum*, which normally creates a carpet of orange flowers during peak bloom, produced very few capitula (inflorescences) in 1986, 1988, and 1992. Thus, newly emerged bees beginning their first nests may have been faced with a dearth of floral resources. *M. inermis* numbers were not affected as strongly at the F2 site in 1988 because of a substantial population of *Centaurea maculosa* that bloomed in late July, in spite of the drought and hot temperatures. This plant was not as abundant at the F2 site in 1986. It is absent from the CL and F1 sites, and was only found in low numbers at the C5 site in 1988. In 1992, cold temperatures through out most of the season also kept nest production low.

Cumulative numbers of nests constructed over the season at the four sites are presented by year in Fig. 14. Final nest numbers are underestimates for *M. inermis* in 1985, as explained above. There are differences between sites and years in dates of first and last nest construction, and in rates of nest construction through the season. *M. inermis* generally started nesting later than *M. relativa*. Both species began nesting earlier in June in 1987 than other years. *M. relativa* began nesting later in 1985, 1989 and 1990 than in other years. Accumulation of nests was slower at most sites during the drought years of 1986 and 1988 than for other years for *M. inermis*. This was not true for *M. relativa*.

The midpoint of the season varied between years, sites and species. Vertical lines on Fig. 14 indicate the date on which 50% of the nests had been started for each species and year. This date was the last date on which nests were classified as early season nests. Early season ended later for *M. inermis* than for *M. relativa* in all years except 1988.

Hypothesis 1: The average length of cells for each offspring, and/or the average number of cells produced per nest is unchanged by exposure to ELF electromagnetic fields.

M. relativa

Mean cell length was calculated for each nest, and used in GLM analysis. If ELF EM fields have an effect on cell lengths we would expect to see mean cell lengths changing for the treatment sites but not for the control sites starting in 1989. This does not seem to be the case. There are no consistent trends of differences between experimental and control areas, either in pre-operational years, or under full power in 1989-91 (Fig. 15). Indeed, the means for control and experimental sites overlap considerably both before and after the antenna became operational. Dashed horizontal lines bracketing the means for each year in Fig. 15 indicate upper and lower limits to the minimum detectable differences between control and experimental means for that year. Difference between actual means was always less. GLM analysis (Table 5) confirms that Exp does not contribute significantly to variation in mean cell length. However, Exp * Antenna is significant at the $\alpha = 0.05$ level both with and without expected sex included in the model (Tables 5,6).

We do not have a biological explanation for the small but statistically significant ($P=0.0244$) Exp*Antenna interaction. The magnitude of the interaction is 0.1mm, indicating that the change in mean length in the experimental sites differs from

the change in mean length for the control sites by only 0.1mm. This statistical result could be a false positive given the many comparisons made in the study. The power of the test for the Exp * Antenna interaction, despite the large sample size, is only 0.61 - 0.72 for $\alpha = 0.05$ (Table 5,6). This means that there is as much as a 39% chance that the interaction was declared significant when in fact it is not significant, i.e., not a very reliable result. In contrast, effects that do have biological meaning such as expected sex (females 0.6mm > males), site (CL=0.25mm larger than other sites), year (1985 = 0.47mm larger than other years), and season (early season cells = 0.33mm larger than late season cells) are highly significant ($P < 0.0001$, Tables 5,6). Compared with these large sources of variability, the small effect of ELF EM fields, if real, is not likely to have a detrimental impact on bee populations.

Overall, the mean cell length per nest was 11.6mm for *M. relativa* (Table 5). The model accounted for only 16% of the variance in mean cell lengths (see r^2 in Table 5) without expected sex included in the model. When expected sex is included, the model accounts for 27% of the variance (Table 6). Female cells averaged 0.6mm larger than male cells (11.55 ± 0.90 mm vs. 10.90 ± 0.87 mm). Between nest variability (error ms) is large. Cell lengths decreased slightly as diameter increased. In addition, cell length decreased as the number of cells in a nest increased. This may reflect in part a decrease in cell length as cells get closer to the nest entrance. Nests with few cells have large inner cells, so mean cell length is large. Nests with many cells include small cells near the nest entrance, so mean cell length will be lower than for nests with few cells. Cell lengths in early season nests tend to be larger than cells in late season nests. This is contrary to the effect of cells per nest, because early season nests tend to have more cells per nest than do late season nests.

In summary, *M. relativa* cells may increase in length in response to ELF EM fields, but the magnitude of the change is so small that it is unlikely to have a detrimental effect on bee populations.

Within and Between Measurer Variability. As mentioned earlier, differences between measurers usually contributed to the variance in mean cell lengths. Mean cell lengths for individual measurers varied from 10.5mm (BZ, 1991) to 11.7mm (VS, 1983) (Table 7). The range of means between measurers was greatest for 1986 cells, when four measurers were involved (0.6mm). It decreased considerably for 1987 cells (0.1mm), but increased again for 1988 - 1991 cells (0.4-0.5mm).

In order to better understand the contribution of measurer differences to cell length variability, in 1987 39 *M. relativa* cells were measured up to three times by each measurer after the cell was originally measured. In an initial two-way model II ANOVA there was no significant interaction between measurers and cell measured. This indicates that although the mean cell length differed between measurers, the magnitude of the differences between cells was the same for all measurers.

The interaction and error variances were pooled by rerunning the ANOVA without including the interaction term in the model. This omission had the additional advantage that the residuals from the model were normally distributed, whereas with the interaction term the residuals were not normally distributed. Each person measured each cell an average of 2.55 times; this value was used to compute the relative contribution of within- and between- measurer variance to the total variance (Table 8). 75% of the variance was between cells, while only 25% was between and within measurers. Variance within measurers (15%) accounted for more of the measurer variance than did variance between measurers (10%). In this analysis the mean cell length was 10.5mm, and the overall coefficient of variation was only 3.6%, or a standard deviation of 0.4mm. Measurer variance accounts for 25% of the total variance, and thus 50% or the standard deviation = 0.2mm. In the full analysis, the overall mean cell length was 11.1mm with a CV of 7.6%, or a standard deviation of about 0.8mm. Thus, our analysis suggests that measurer variance accounts for about 0.4mm of the total variance.

M. inermis

As with *M. relativa* nests, mean cell length was calculated for each nest and used in GLM analysis. However, in some years, numbers of nests are very low at some sites (eg., 1 nest with female cells at the CL site in 1986 and 1988). Therefore, cell length data were pooled for 1985-1986 and for 1987-1988. Only nests with diameters greater than 9.5mm were used in the analysis. The residuals were significantly different from normal in this analysis (Kolmogorov D = 0.017 N=3070, P = 0.024), but a histogram of the residuals appeared to be very close to normal, so the GLM results should be accurate.

If ELF EM fields have an effect on *M. inermis* cell lengths we would expect to see mean cell lengths changing consistently for the treatment sites but not for the control sites as ELF EM fields increase. This does not seem to be the case (Fig. 16). GLM analysis (Table 9) confirms that neither Exp nor Exp*Year contribute significantly to variation in mean cell length.

Therefore, there does not appear to be any influence of ELF EM fields on cell length for this species.

M. inermis cells expected to have female offspring averaged 1.2mm larger than cells expected to have male offspring (16.3mm - 15.2mm). The model accounts for 46% of the variance in mean cell lengths. Parameters that contributed significantly to *M. inermis* cell length were similar to those that were significant for *M. relativa* cell length. Cells from the CL site tended to be significantly larger than cells from C5, and cells from F1 were larger than cells from F2. Cell lengths decreased slightly as number of cells in a nest increased. Cells in complete nests tended to be larger than cells in incomplete nests.

As with *M. relativa*, differences between measurers (Measurer [year * Antenna]) made a significant contribution to cell lengths. Cells measured by KS were larger than cells measured by other measurers (Table 10). In 1990 and 1991, 39 *M. inermis* cells were measured three times by each measurer after the cell was originally measured. A two-way random-effects model ANOVA will be used, as for *M. relativa*, to partition the variance within and between measurers and between cells. We have not completed these analyses as of this writing.

In summary, *M. inermis* cell lengths are not affected by ELF EM fields.

Offspring Weights. In the 1987 Annual Report we questioned the necessity to analyze the variance in cell volumes, because volumes are highly correlated with nest diameters. We suggested that the answer to this question depended on whether offspring weights correlate best with cell length or with cell volume. In last year's annual report we showed that bee weight is unrelated to cell length or volume, and thus does not help us resolve the question of whether we should be analyzing variance in cell volume rather than cell length. Since the two measures are correlated, and since our analyses have thus far been on cell length, we decided to continue to analyze only cell length in the future.

However, we should be able to do a separate GLM analysis of bee weight as a dependent variable, just as we have analyzed cell length, for 1987-1990 bees. Such an analysis has not yet been done. It will add an additional hypothesis to our study: **Newly emerged bees exposed to ELF EM fields are the same weight as newly emerged bees not exposed to ELF EM fields.** We hope to be able to add this hypothesis to the final version of this annual report. Both live and dry weights of a sample of both bee species from 1987 - 1991 nests have been measured. Data

We hope to have a GLM analysis completed for the next annual report.

After weighing, the bees were pinned and identified. All of the small *Megachile* bees from 1986 - 1988 nests have been confirmed as *M. relativa*. The 1989 - 1991 bees will be identified after they are weighed and pinned.

Number of cells per nest

Number of cells per complete nest ranged from 1 to 12 for *M. relativa*. In a CATMOD analysis of cells per nest we used four categories to minimize the cases in which expected frequency was less than five. The categories were: nests with 1 or 2 cells, nests with 3 or 4 cells, nests with 5 or 6 cells, and nests with seven or more cells (Fig. 19).

There were significant differences in the distribution of number of cells per nest between Sites, Years, and Exp areas. However, the interaction between Exp and Antenna was not significant (Table 14), indicating that none of the variability in cells per nest for *M. relativa* can be attributed to antenna operations at the experimental sites.

Similar patterns can be observed when early vs. late season nests are included in a CATMOD analysis. Two categories of nests were used: 1-4 cells, and 5-12 cells (Fig. 20, Table 15). This analysis indicates that late season nests usually have fewer cells than early season nests. Differences between years, and sites are also significant. However, the Exp * Antenna interaction is not significant, indicating that none of the variability can be attributed to antenna operations at the experimental sites.

Number of cells per complete nest ranged from 1 to 8 for *M. inermis*. The deeper the nest, the more cells can be constructed. Therefore, in analyzing cells per nest for *M. inermis*, we compare only 1987 - 1991 nests, when bore depth was routinely 140mm and only drill bits of 11mm were used to make large diameter nests. In all years, the experimental sites have more cells per nest than do control sites (Fig. 21), as confirmed by CATMOD analysis (Table 16) using two categories (1-4 cells or 5-7 cells). No significant Exp * Antenna interaction indicates no effect of ELF EM fields at experimental sites after 1989. The significant "Antenna" effect means that cells per nest differed in 1987 - 1988 as compared with 1989 - 1991 at all sites. Thus, these differences have nothing to do with ELF EM fields.

Early vs. late season was added to the catmod analysis for *M. inermis* (Fig. 22, Table 17). In this analysis, nests from

1987 and 1988 were pooled because of small sample sizes. Season was significant, with more large nests produced early season than late season. Exp was significant, presumably because nests from the experimental sites, F1 and F2, had a greater proportion of large nests than do nests from control sites. Year*season was significant, suggesting that some year-season combinations have a greater proportion of small nests than do other year-season combinations. The Exp*Year and Exp*Year*Season interactions were not significant, indicating that the fully operational ELF EM fields in 1989 - 1991 did not affect early or late season nests differentially.

In summary, the number of cells per nest is unchanged by exposure to ELF EM fields.

Hypothesis 2. Bees exposed to ELF EM fields, and bees not exposed, will make nest plugs of the same thickness and will devote the same proportion of nest space to reproduction.

We have at last begun an analysis of nest plug length for *M. inermis*. In a GLM analysis similar to those performed on cell lengths, we found that residuals were significantly different from normal (Kolmogorov D = 0.096, N=2307, P < 0.01). A histogram of the residuals suggests that the distribution is more leptokurtic than a normal distribution. We have not determined a transformation that would adjust for this problem, so we have used a GLM of the ranks of nest plug lengths in a non-parametric factorial analysis of the data (Zar p. 250). The results of this analysis were qualitatively identical to the results of the GLM on the raw data. We present the results from the later analysis in Table 18.

If ELF EM fields have an effect on *M. inermis* nest plug lengths we would expect to see nest plug lengths changing consistently for the treatment sites but not for the control sites as ELF EM fields increase. This does not seem to be the case. Control sites tend to have larger nest plugs than experimental sites during most years (Fig. 23). GLM analysis confirms that Exp is significant, but the Exp*Antenna interaction is not significant. Therefore, there does not appear to be any influence of ELF EM fields on nest plug lengths for this species.

Not surprisingly, cells per nest accounts for the most important contribution to variance in nest plug lengths. Nest plugs average over 6 mm less in length as each new cell is added to the nest, presumably because there is less space available in the nest for plug. Nest plug lengths also increase significantly as diameter increases, and are larger in early season nests. Plugs at the control sites average about 3 mm longer than at experimental sites. Plugs were shorter in

1985 and 1986, presumably because most trap nests were shorter and had fewer cells than in subsequent years (see methods).

In summary, nest plug lengths do not change in response to ELF EM fields.

Hypothesis 3. The number of leaves used to line a cell is unchanged when bees are exposed to ELF EM fields.

Although the number of leaves lining a cell is discrete data, we treat these data as if they were continuous, and use the GLM procedure instead of a CATMOD analysis. This should increase our ability to detect differences between control and experimental areas if any exist.

A mean of ln leaves per cell was calculated for each nest and used in GLM analysis. For *M. relativa*, analysis starts with 1986 data, when leaves were first counted. For *M. inermis*, numbers of nests are very low in some years at some sites (eg., 1 nest with female cells at the CL site in 1986, 1988). Therefore, data on leaves per cell were pooled for pre-operational years 1985-1986 and 1987-1988. Only nests with diameters greater than 9.5mm were used in the *M. inermis* analysis. The residuals were significantly different from normal in the *M. inermis* analysis (Kolmogorov D = 0.018 N=2889, P = 0.021), but a histogram of the residuals appeared to be very close to normal, so the GLM results should be accurate. If the 1990 data are deleted from the analysis, the residuals are normal.

If ELF EM fields are having an effect on leaves per cell, we would expect to see mean leaves per cell changing for the treatment sites but not for the control sites as EM fields increase. For *M. relativa*, there were fewer leaves per cell in 1986 and 1987 (Fig. 24) than in subsequent years. However, the magnitude of the changes between off and on antenna years was not significantly different for control and experimental sites (Exp*Antenna F=0.07 df=1 P=0.80; Table 19). The same is true if expected sex was added to the model (Exp*Antenna F=0.06 df=1 P=0.20 Table 20). Thus, ELF EM fields do not have an effect on leaves per nest for the small bee species.

For *M. inermis*, leaves per cell appears to increase in 1990 and 1991 compared with earlier years (Fig. 25). GLM analysis (Table 21) indicates that the Exp*Antenna interaction is significant for leaves per cell (F=7.46, df=1, P=0.0063). Control cells increased about .5 leaves between off and on years (12.5 increasing to 13.1 leaves per cell), while experimental cells increased about 1.4 leaves between off and on years (11.8 increasing to 13.2 leaves per cell). The control

and experimental sites differed more before the antenna became operational than they did after the antenna became operational. This is the reverse of the pattern that we would like to see to explain a significant Exp*Antenna interaction, namely the control and experimental sites being the same before the antenna was operational, and the experimental site changing only after the antenna became operational. The actual results suggest that control sites intrinsically have more leaves per cell than experimental sites, as was apparently the case in 1985-1988, and that *M. inermis* may be padding its cells with an extra leaf in the presence of ELF EM fields, so that now there is no difference between experimental and control areas.

Unfortunately, sample size for *M. inermis* is smallest for the "off" antenna years of 1985-1988, on which the inference of intrinsic differences between control and experimental sites is based. The power of the GLM test is 0.78 for $\alpha = 0.05$, i.e., there is a 22% chance that this result is a false positive. We would feel more confident that the significant interaction was caused by ELF EM fields if bees were padding their nests in other ways as well. No evidence of such padding was seen in our analysis of *M. inermis* nest plugs.

Despite these caveats, let us assume for the moment that bees do increase the leaves per cell by one leaf in response to ELF EM fields. What difference does this make? It takes an average of about 2.4 minutes to collect a leaf and about 2.8 minutes to position it in the nest (Strickler, unpublished data). If there are 8 cells in a nest (a maximum), this adds about 40 minutes to the week or so required to make a nest. The bee population is able to accommodate considerable variability in leaves per cell, and in the time to collect leaves, due to other factors. These include offspring sex (13.3 leaves for male cells, 11.6 leaves for female cells; $P < 0.0001$, Table 21), diameter (about 1.2 leaves per mm; $P < 0.0001$, Table 21), and intrinsic differences between nests (reflected in the high Error MS = 89.61 and low $r^2 = 0.37$; Table 21). Thus, we expect that an additional leaf will have little impact on the total reproductive output of a bee. Our finding of no significant Exp*Antenna interaction in cells per nest for *M. inermis* is consistent with this expectation.

However, the brief additional time out of the nest may increase the risk of exposure of the cell to parasites, to predation or other mortality factors outside of the nest, and/or to usurpation by other species. Risk of parasitism from flies in the genus *Anthrax* spp. is increased by the absence of a female bee at the nest when the cell is being constructed because these parasites flip eggs indiscriminately into unguarded openings (Scott and Strickler, 1992). In contrast, cuckoo bees of the genus *Coelioxys* spp. tend to lay their eggs

just after the *Megachile* lays her egg, not when the cell is being constructed). We used a CATMOD analysis to compare the number of cells containing *Anthrax* parasites to the number of cells with other emergences. There were significant differences between sites and years for *M. inermis* (Site[Exp] $\chi^2=14.19$ df=2 P=0.0008; Year[Antenna] $\chi^2=14.16$ df=5 P=0.0146), but no significant Exp*Antenna interaction ($\chi^2=0.14$ df=1 P=0.7115). Thus, we have no evidence that parasitism increases for *M. inermis* as a result of the addition of an extra leaf per cell in nests constructed under the operational ELF antenna.

There is no direct way to measure predation of bees foraging for leaves, but we can compare the number of incomplete and usurped nests as an indirect measure of such predation. Interestingly, incomplete *M. inermis* nests have significantly more leaves per cell than do complete nests (Table 21; "incomplete" nests includes usurped nests in our analyses). Leaves per cell do not differ for such nests in *M. relativa* (Tables 19, 20). These results are consistent with the possibility that an additional leaf is the cause of the incomplete nests for *M. inermis*. In a CATMOD analysis of complete vs. incomplete nests, there is a significant Exp*Antenna interaction ($\chi^2=6.01$ df=1 P=0.0142) for *M. inermis*. However, the significant interaction is caused by a slight reduction in incomplete nests at experimental sites with antenna operational (Fig. 26). This is the reverse of the effect expected from an increase in leaves per cell.

Thus, we conclude that the ELF EM fields may be causing *M. inermis* to pad its cells with an average of one extra leaf, but there is no evidence that this is having a detrimental effect on the bee population.

Hypothesis 4. The relative acceptability of nests oriented in a NS direction vs. nests oriented in an EW direction does not change when bees are exposed to ELF EM fields.

As explained in the methods section, at each site there are three sets of hutches. Each hutch set consists of two hutches in close proximity, one oriented N-S, and one oriented E-W. Nests on the N-S hutch have openings facing E or W, while nests on the E-W hutch have openings facing N or S. The directions used in this analysis refer to the direction of nest openings.

Each set of hutches is situated in a different location and has a different pattern of sun and shade during the day, and a different compliment of nearby flowering plants. These factors may be important in acceptance of nest opening direc-

tion by bees. Thus, we have analyzed nest orientation by hutch set at each site. Furthermore, since sample sizes are low at some hutches in some years, we have not tried to discriminate between nests oriented in four directions; rather we compare acceptance of nests oriented N or S vs. nests oriented E or W. Only data for *M. relativa* are analyzed, since sample size was very low most years for *M. inermis* at the control sites.

We analyzed the data with a Log-likelihood Ratio (G-test) Contingency test (Table 22). This tests whether the pattern of nest acceptability (whatever the pattern) is the same for all years at a given hutch set. When the null hypothesis was accepted for all hutch sets at a site the data were pooled over years and each hutch set was tested against the other hutch sets at that site, to test whether the pattern was consistent for the entire site.

If ELF EM fields affect nest orientation acceptability, one would expect changes in nest orientation within a hutch set over the years at experimental but not control areas. The results indicate that at three of the four sites there is a consistent bias over the years at a given hutch set, but that the bias is different between hutch sets at a given site. These biases are probably due to differences in shading and proximity to resources, which are fairly consistent between years. Only at F1 have two hutch sets shown significant changes within a hutch set over the years. For F1-N, these differences appear to be due to differences between 1985 and subsequent years. If a G-test is repeated with 1985 data removed, the F1-N hutch has a consistent bias (3:10) toward the NS direction ($G=6.439$, $df=5$ n.s.). We have no idea why nest directions were different in 1985 than in subsequent years at the F1-N hutch, but this change cannot be related to ELF EM fields. Similarly, nest orientations at the F1-W site have changed in both pre-operational years (eg, 1983 vs 1985, 1985 vs 1986, 1988) and full operational years (1990 vs. 1991). Thus, these changes can't be attributed to ELF EM fields.

Last year the F2-N hutch set showed a change in nest orientation in 1989 that continued in 1990, both full power years. We argued that this change was not likely to be attributable to ELF EM fields. However, when 1991 distributions are added to the analysis, we were able to accept the null hypothesis that there is a bias (4:3) toward EW nests at this hutch during all the years that the hutches have been in their current location (1987-1991).

In summary, nest orientations do not change when bees are exposed to ELF EM fields.

V. NEST ACTIVITY RESULTS

Hypothesis 5. The duration of round leaf (LO) foraging trips remains the same when bees are exposed to ELF EM fields.

No new data were collected for this hypothesis in 1992. However, the analysis of the data has been updated to be consistent with other models in this report.

During the 1987 field season we noticed that LO trip durations increased with each successive trip after the bee lays her egg. In 1987, however, we did not keep track of which LO trips in the capping sequence were being timed. However, we learned that the female makes a series of very rapid flights in and out of the nest just before collecting the first LO after laying her egg. Undergraduate observers refer to this behavior as "spazzing". Where rapid flights in and out of the nest, without a cargo, appear at the beginning of a series of 1987 LO timings, we have assumed that the first LO trip for the cell has been timed.

In 1988 we recorded the actual trip number for 73% of the capping sequences that were timed. In 1989 - 1991, we were even more diligent, recording actual trip numbers for every cell cap timed. In our analyses residuals fit a normal distribution when we restrict the analysis to the mean of the first 3 trips if we use a $\ln(\ln)$ transformation of LO trip durations.

Figure 27 summarizes mean LO durations for the four sites and five years, based on GLM analysis of the mean of trip ranks 1-3 for each cell capping bout. If ELF EM fields were having an effect on LO durations, we would expect to see mean durations increasing (or possibly decreasing) for the treatment sites but not for the control sites since 1989. This does not seem to be the case. LO durations tend to be greater at the experimental sites across all years, even before the antenna became fully operational. Furthermore, mean LO durations have tended to fluctuate around a narrow range of means from year to year. (There was a greater spread between sites in 1987 than in subsequent years because of smaller sample sizes.) Results of the GLM analysis of the mean of trips 1-3 are summarized in Table 23. The error variance is a measure of between bee variability. There is a significant Exp effect, but the interaction between Exp and Antenna does not contribute significantly to the variability. These results indicate that ELF EM fields do not affect LO trip durations.

Time of day did not contribute significantly to variability in LO durations. This suggests that temperature or other

weather parameters do not affect LO durations. Date of the timing was significant only in 1990.

Although 1991 was the final year for collecting data on LO durations, there are still several refinements to the analysis that we plan to make to confirm the results presented here. First, we are in the process of analyzing data collected prior to 1987 using the same GLM procedures, to see if 1983-1986 data are consistent with "off" antenna results in 1987-88. At the time of this writing, the Trip Rank variable is being created for these data. Second, we hope to include weather as a covariate in the final analysis.

VI EMERGENCE RESULTS

We have noted that the expected sex of an offspring contributes significantly to variability in cell lengths and leaves per nest, and we have argued (p.7) that changes in sex ratio could occur as a result of stress from ELF EM fields. It is appropriate, therefore, to consider whether there have been any significant changes in sex ratio that can be attributed to ELF EM fields. In this report we test a new hypothesis: **The relative proportions of emerging males and females is unchanged by exposure to ELF EM fields.**

We have tested this hypothesis with *M. relativa* sex ratio. *M. inermis* data are not suitable for testing this hypothesis because there were changes in nest diameters and depths in 1987 that could affect sex ratio (Stephen and Osgood, 1965). Table 24 presents numbers of males and females emerging, and sex ratios, for each site and year since 1985 for *M. relativa*. Figure 28 graphs the relative proportions of each sex. There has been much variability between sites and years. Results of a CATMOD analysis of the frequencies of each sex of (Table 25) indicate that there are significant differences between experimental and control areas and between years in the relative frequencies of the sexes. However, the Exp * Antenna interaction is not significant, so none of the variability can be attributed to ELF EM fields.

Hypothesis 6. Overwintering mortality of megachilid bees is unchanged by exposure to ELF EM fields.

For the most part, both species of *Megachile* in our study are univoltine, having only one generation per year. There have been a few exceptions: In *M. relativa* nests, 5 - 22% of all *M. relativa* and 6 - 26% of all *Coelioxys* spp. emergences occur in August and September (Table 26). Far fewer instances of bivoltinism occur in *M. inermis* nests (0 - 0.3%; Table 27). Early emergences do not overwinter, and are not included in the analysis described below.

Prior to emergence as an adult in the spring, *Megachile* are subject to a variety of sources of mortality. The egg may fail to hatch, or the larva may die of unknown causes during the summer. The prepupa may die during the winter. The pupa may fail to eclose in the spring. A number of parasites may attack the *Megachile* egg, larva, or pupa at various times in its development. Parasites include the cuckoo bees, *Coelioxys moesta* Cresson on *M. relativa* and *C. funeraria* Smith on both *Megachile* spp.; the flies *Anthrax irroratus irroratus* Say and *Anthrax pluto pluto* Weidemann; chalcid and leucospidid wasps.

The percent mortality due to various causes is presented by site and year for *M. relativa* and *M. inermis* in Tables 28 and 29. Variability between years is due in part to a change in protocol in 1987, leaving nests to overwinter in the field rather than bringing them to Channing. For example, Pre-overwintering mortality (mortality of eggs and larvae) was greater in 1987 than in previous years, and even greater in 1988, especially for *M. relativa*. Weather patterns are undoubtedly also involved. High pre-overwintering mortality in 1988 nests was probably due to dry, hot summer weather. Unusually cold spring weather contributed to overwintering mortality of nests constructed in 1988 and 1989. Numerous summer rainfalls may have caused higher pre-overwintering mortality in 1987 as compared to earlier years. Similarly, proportion of adults emerging was particularly low for 1988 *M. relativa* nests.

There are several ways that one can measure overwintering mortality, and several problems that must be dealt with in analyzing it. First, we equate overwintering mortality with the prepupal stage, but actually the prepupa lasts for a longer time than just the winter. The prepupal stage begins several weeks after the egg is laid, when the larva has finished eating its provisions. The prepupa defecates shortly after molting, and then spins a silken cocoon for overwintering that is surrounded by fecal pellets. Thus the prepupal stage may begin as early as mid-summer. It lasts until pupation in the spring. This occurs typically in mid May to mid June, although we have opened few cells to find out, to minimize mortality. In spring 1989 and 1990, the prepupal stage for nests constructed in 1988 and 1989 probably lasted into late June, due to cool weather and a change in protocol to a shady outdoor emergence site. Emergence was delayed in 1989 - 1991 until July. Figs. 29 - 33 compare emergence of 1987 - 1991 nests in spring of 1988 - 1992. Prior to 1989, pupation and emergence took place in the lab where indoor microclimate and 60 hz EM fields could affect pupal and adult mortality. Starting in 1989, the effects of 60 hz EM fields were minimized by moving emergence of all cells to an outdoor holding site.

There is no way to separate prepupal mortality that occurs during the winter from prepupal mortality that occurs in summer, fall or spring. 1987 - 1989 nests were left at the sites where they were constructed during the entire prepupal stage except for the last few weeks, when nests were returned to Crystal Falls for nest architecture measurements. Thus, the effects of ELF EM fields on prepupal mortality any time before May are tested by our protocol.

We have no way of knowing how many adult bees would have successfully emerged at the study sites, but the number of cells that survive past the prepupal stage provides an upper

limit. Therefore, we combine pupae, adults that die in the cocoon, and adults that successfully emerge, into one "post-overwintering" category.

The prepupal stage has the longest duration of all the developmental stages of these univoltine species. However, mortality is usually greater in the pre-overwintering egg and larval stages. Mortality of these early stages show intrinsic differences between sites and differences between years that are weather related (Tables 28, 29), that could make it difficult to detect differences due to ELF EM fields. Therefore, we propose restating our hypothesis as: **Given that a bee survives to the prepupal stage, the probability that it will not survive past the prepupal stage does not change in the presence of ELF EM fields.** Thus, we compare the number of cells with a dead prepupa with the number of cells with post-overwintering bees. Cells containing egg and larval mortality are not included in the analysis.

Parasites present another problem. It is easy to distinguish adult and pupal *Megachile* from adult and pupal parasites. However, we are unable to distinguish prepupae of *Megachile* from prepupae of the cuckoo bee, *Coelioxys* (also in the Megachilidae). The *Coelioxys* larva kills its host larva or egg, and feeds on the provisions in the cell. Like the host bee, *Coelioxys* overwinters in the prepupal stage. When testing the hypothesis above, the number of cells with dead prepupae should be reduced by the percentage of cells that are parasitized by *Coelioxys*. We can estimate percent parasitism of prepupae from the proportion of adults that are parasites. This assumes that there is no differential mortality of parasites in the prepupal stage as compared with the adult stage.

In our attempts to analyze prepupal mortality, however, we have not tried to separate *Megachile* and *Coelioxys* data. Rather, we assume that both genera are affected in the same ways, if at all, by ELF EM fields. This assumption is more likely to be true for two bee species in the megachilid family, than for a bee and a fly or wasp parasite. The percent of cells with prepupal mortality for each site and year (number of cells containing dead prepupae divided by all cells with *Megachile* or *Coelioxys* prepupae or post-overwintering stages x 100) is graphed in Figs. 34 and 36. A Tukey test of arcsine transformed proportion of prepupal mortality was used to test for differences between years. Years with the same letter above the graphs in Figs. 34-37 were not significantly different.

In 1989 nests, prepupal mortality often occurred in several cells in a row in a nest. Some of these cells had a partially formed pupa visible under the prepupa exoskeleton.

These prepupae obviously died late in their development, just before pupation. We believe this occurred during the cold spring weather, particularly on May 10, when there was a snow storm. Emergence of each sex in a nest tends to occur within two or three days, although emergence of the entire population takes much longer. This suggests that development is synchronous within a nest, so that mortality at critical stages of development may be autocorrelated. For 1989 nests in particular, prepupal mortality in a cell was probably not independent of prepupal mortality of other cells in the same nest, which were all at the same stage of development when cold weather occurred. Therefore, in addition to an analysis of prepupal mortality by cells, we have analyzed prepupal mortality by nest. Percent of nests with prepupal mortality for each site and year (number of nests containing at least one dead prepupa divided by all nests with at least one *Megachile* or *Coelioxys* prepupa or post-overwintering stage $\times 100$; Fig. 35, 37) was higher than the percent of cells with prepupal mortality (Figs. 34, 36).

1985 and 1986 nests were not overwintered at the sites where they were constructed. Therefore, analyses of prepupal mortality include nests constructed between 1987 and 1991. This analysis includes 2 "off" antenna years and 3 "on" antenna years. Results are presented in Tables 30-33.

Exp*Antenna did not contribute significantly to variance in proportion of cells or nests with prepupal mortality for *M. relativa* (Tables 30, 31). This suggests that exposure to ELF EM fields during the winters of 1989-90, 1990-91, and 1991-92 did not affect overwintering mortality. Year was significant in all tests, as noted in Figs. 34-35, but the differences do not correspond to the operational status of the ELF antenna.

The Exp * Antenna term is significant for *M. inermis* cells and nests (Tables 32, 33). Examination of Figs. 36 and 37 suggests that the percent of cells or nests with prepupal mortality increased more at the experimental sites in 1989 and 1990 than at the control sites. The pattern is less apparent in 1991.

In the manipulative experiment, nests (and cells) constructed at the F2 site but overwintered at the C5 site, had mortality closer to nests constructed and overwintered at C5 than to nests constructed and overwintered at F2 (Fig. 38, Table 34). This indicates that winter conditions at the F2 experimental site cause greater prepupal mortality than do conditions at the C5 control site. One such condition may be exposure to ELF EM fields. This is consistent with the observation that both cell and nest mortality were greater at C5 than at F2 during pre-operational years of 1987 and 1988 (Fig. 36, 37). The reverse has been the case for the opera-

tional years of 1989-1991. The result is also consistent with the significant Exp * Antenna interaction in our analysis of 1987 - 1991 *M. inermis* prepupal mortality (Table 32, 33).

In summary, we have evidence that *M. inermis* but not *M. relativa* overwintering mortality may be affected by ELF EM fields. This result is based on only two "off" antenna years of data. This summer we will add emergence data for 1992 nests. If the control sites have greater mortality than experimental sites in 1992 as in 1987-1988, then we may alter our conclusion. If the pattern of greater mortality at the experimental sites continues, then we will conclude that ELF EM fields significantly increase mortality of *M. inermis* prepupae. Unlike the possible changes in cell length and leaves per cell noted earlier, this increase in mortality could reduce *M. inermis* populations, especially if the increased mortality persists over time.

VII SUMMARY

Studies of the effects of high voltage transmission lines and magnetic fields in honeybees suggest several ways that solitary megachilid bees might be affected by ELF electromagnetic fields. In particular, honeybees show greater levels of activity, reduced reproductive output, lower overwintering survival and modifications of nest structure in response to high voltage transmission lines. In addition, honeybees can detect magnetic fields and may use them in orientation. ELF EM fields may affect megachilid bees in similar ways.

Megachilid bees are particularly well suited for this study. Their investment per offspring and reproductive output per nest are easy to measure because they provide each offspring with a discrete cell, and because they readily nest in artificial nests. Three types of data have been gathered in past years: nest architecture, nest activity, and emergence/mortality.

Two abundant species at the experimental and control sites, both in the genus *Megachile*, are the focus of our analysis. These species differ in size and degree of sexual dimorphism. Thus, they may be impacted differently by ELF EM fields.

Four hypotheses regarding the impact of ELF EM fields on nest architecture are being tested:

Hypothesis 1: The average length of cells for each offspring, and/or the average number of cells produced per nest is unchanged by exposure to ELF electromagnetic fields.

Hypothesis 2. Bees exposed to ELF EM fields, and bees not exposed, will make nest plugs of the same thickness and will devote the same proportion of nest space to reproduction.

Hypothesis 3. The number of leaves used to line a cell is unchanged by exposure to ELF EM fields.

Hypothesis 4. The relative acceptability of nests oriented in a NS direction vs. nests oriented in an EW direction does not change when bees are exposed to ELF EM fields.

Nest architecture data for *M. relativa* nests constructed in 1983, and *M. relativa* and *M. inermis* nests constructed in 1985-1990 have been analyzed. Cells with female offspring were larger than cells with male offspring. Mean cell lengths were significantly larger at the CL site, in complete nests and early season nests, and in nests with few cells. There was no effect of ELF EM fields at 100% power (1989-1990) on cell length for *M. inermis*. However, *M. relativa* cells may be about

0.1mm larger at experimental sites as a result ELF EM fields. This is a very minor contribution to variability in cell lengths and is not likely to affect the bee population.

Number of cells per nest was significantly less for nests begun late in the season as compared with those begun early in the season. The distribution of numbers of cells per nest varied between years, sites, and treatment areas, but neither species showed any obvious changes at the experimental sites in 1989 - 1991 when the antenna was fully operational.

Nest plugs decrease in length as the number of cells in the nest increases, are longer early in the season, and are longer at the control sites. However, there was no effect of ELF EM fields on nest plug length.

Mean number of leaves per cell was smaller for female cells than for male cells, and increased as nest diameter increased. Nests begun in early season had fewer leaves per cell than did nests constructed late season. There was no effect of ELF EM fields on leaves per cell for *M. relativa*. However, for *M. inermis* about 1 extra leaf per cell was found at Experimental areas after the antenna became operational. No evidence was found that parasitism or percent incomplete nests increased as a result of adding an extra leaf per cell. This change should have little if any detrimental effect on bee populations.

Nest entrance orientation has been consistent over the years for *M. relativa* at control sites, and at the F2 experimental site. Orientation has varied at two of three hutch sets at the F1 experimental site, but the changes are not related to ELF EM fields.

One hypothesis regarding nest activity is being tested:

Hypothesis 5. The duration of round leaf (LO) foraging trips remains the same when bees are exposed to ELF EM fields.

There were significant differences between years and between sites in the duration of LO collecting trips, however, there were no significant changes in LO durations at the experimental areas after the antenna became fully operational in 1989. Thus, ELF EM fields, even at full power, have not had any affect on LO trip durations.

Two hypotheses concerning emergence and mortality data have been tested:

This year for the first time we have statistically tested whether sex ratios of *M. relativa* are affected by ELF EM fields. We have not found any significant effect.

Hypothesis 6. Overwintering survival of megachilid bees is unchanged by exposure to ELF fields.

Overwintering mortality takes place when the bee is in the prepupal stage. Because of the effects of microhabitat and year on pre-overwintering (larval) mortality, it was decided to eliminate cells with this mortality from our analysis. Thus our hypothesis has been restated as: Given that a bee survives to the prepupal stage, the probability that it will not survive past the prepupal stage does not change in the presence of ELF EM fields. We compare the number of cells with a dead prepupa with the number of cells with pupae, dead adult, or emerging adult bees. Mortality of the parasitic cuckoo bee, *Coelioxys*, is included in the analysis, since we cannot distinguish the two bee species until the pupal stage.

M. relativa overwintering mortality is not affected by ELF EM fields. However, *M. inermis* cells and nests with prepupal mortality at experimental sites have increased more since the antenna became operational than have cells and nests with prepupal mortality at control sites. When nests from one experimental site are moved to a control site for the winter, mortality is reduced to the level of the control site. These results suggest ELF EM fields may increase overwintering mortality for *M. inermis*. Overwintering data for 1992 nests will be added to our analysis this summer, and we will be interested to see if increased mortality persists.



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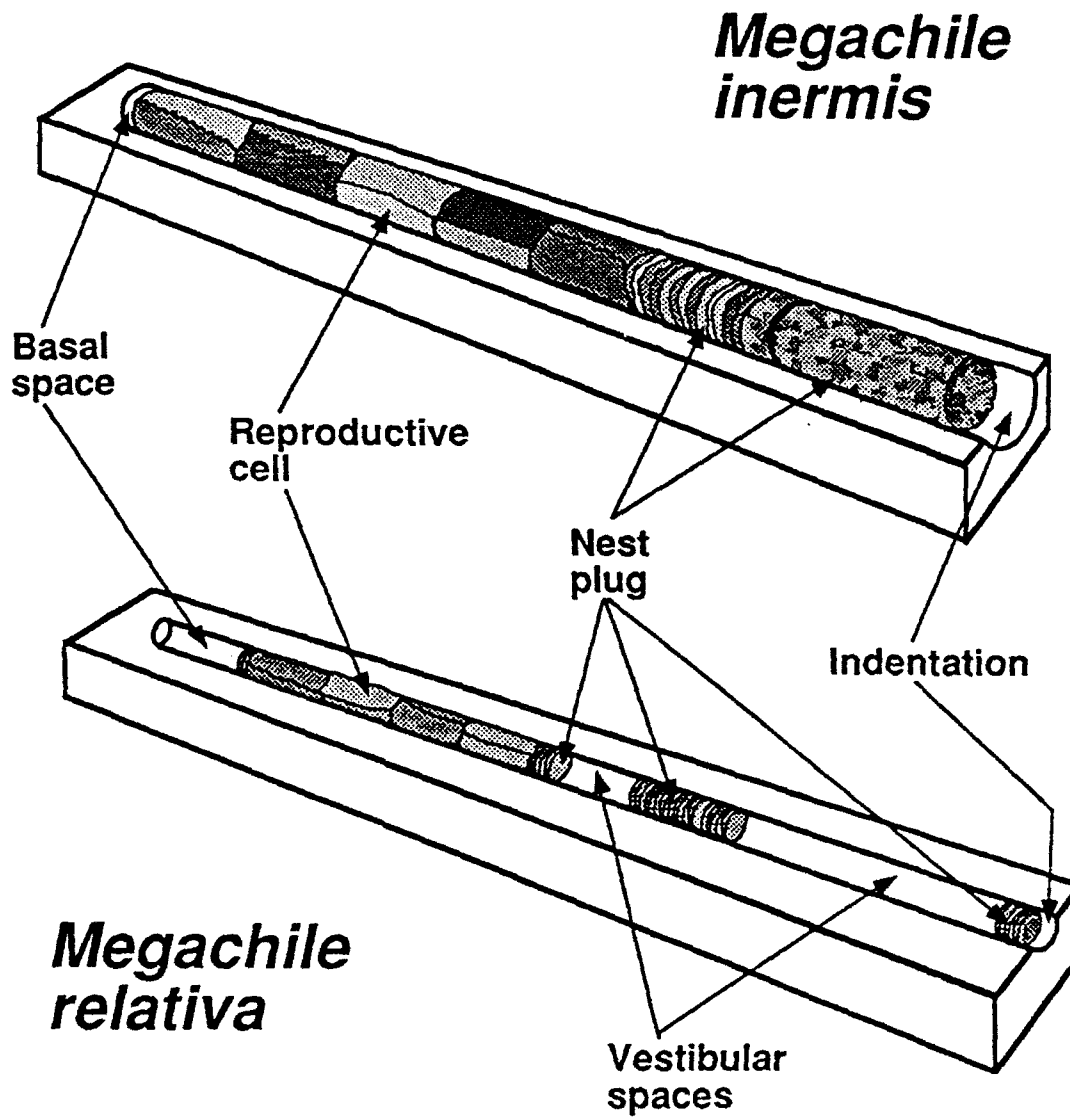


FIGURE 1. Cut away view of a completed *Megachile inermis* and a completed *M. relativa* nest.

TABLE 1: Diameter of drill bits used to create trap nests.

Diameter, mm	Used by <u>M. relativa</u>	Used by <u>M. inermis</u>
4.4*		
5.2*	xx	
5.5	xxx	
6.0	xxx	
7.2*	xx	x
9.4*		xx
11.0		xxx

* Drill bit diameters used before 1987 only.

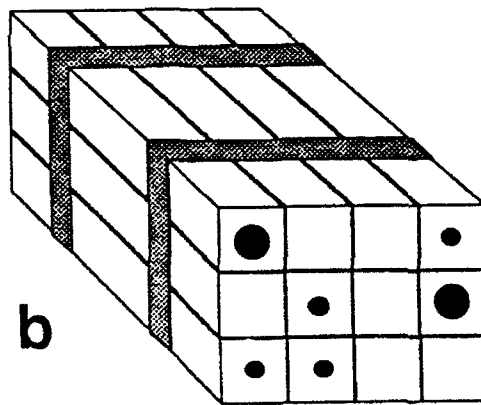
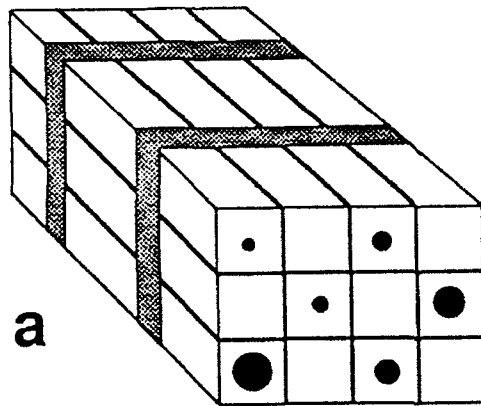


FIGURE 2. Examples of arrangement of nests in a block.
a) 1983-1986 b) 1987-1992

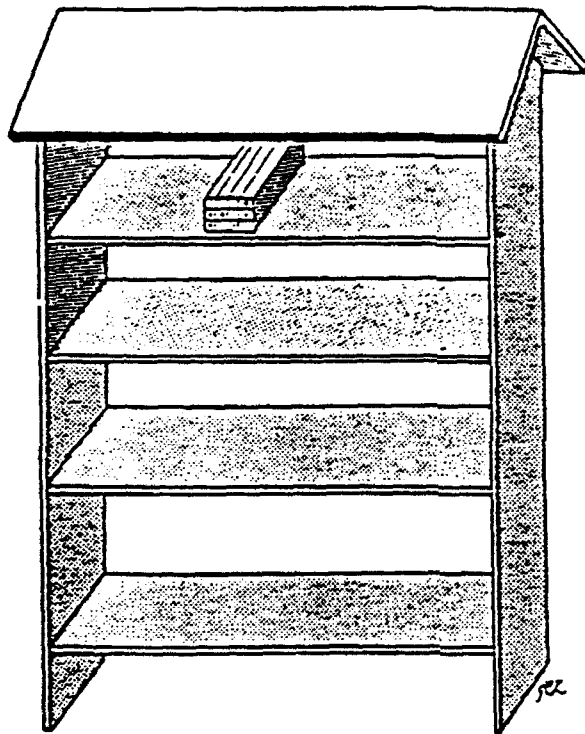


FIGURE 3. Hutch, with one block of nests.

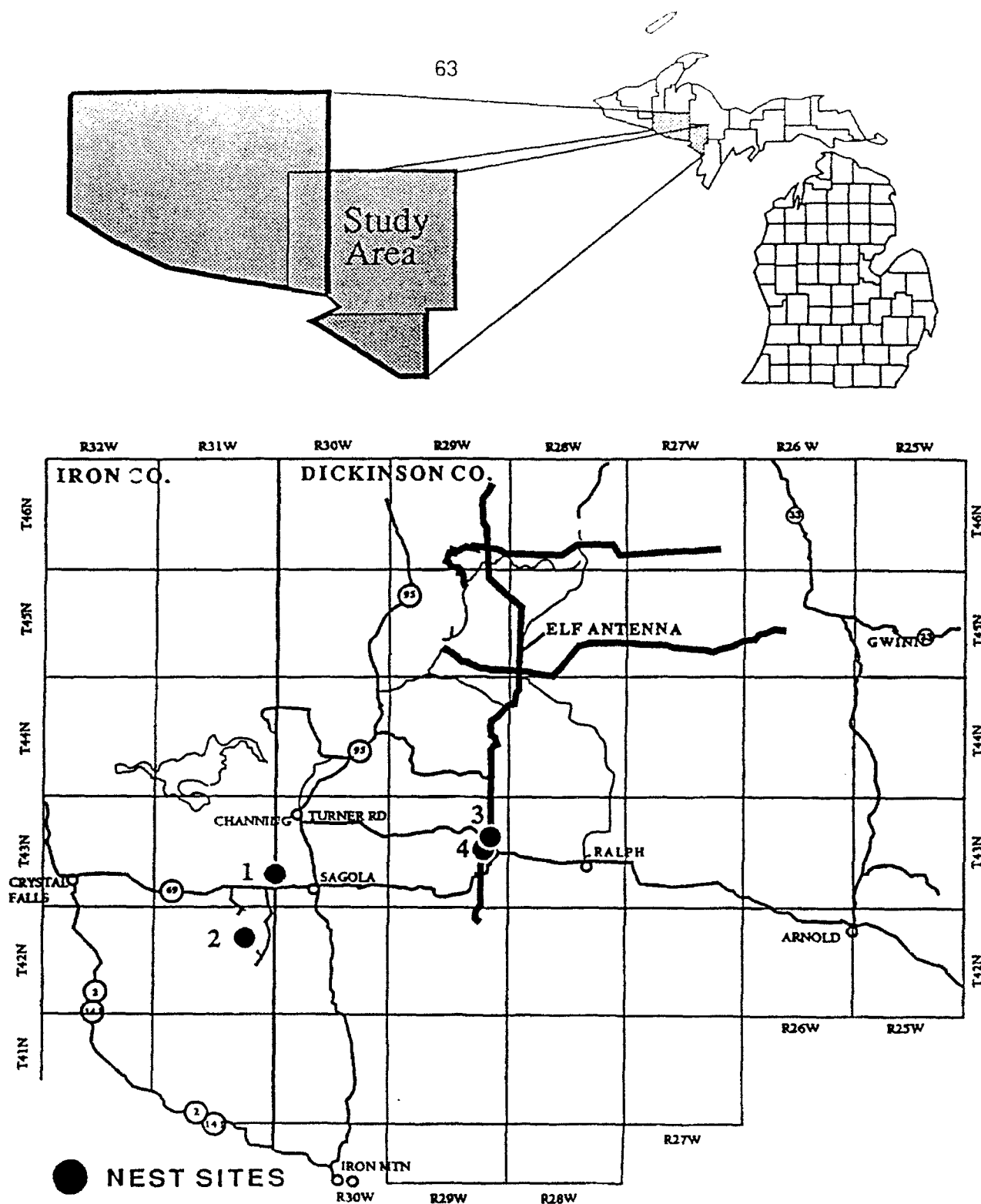
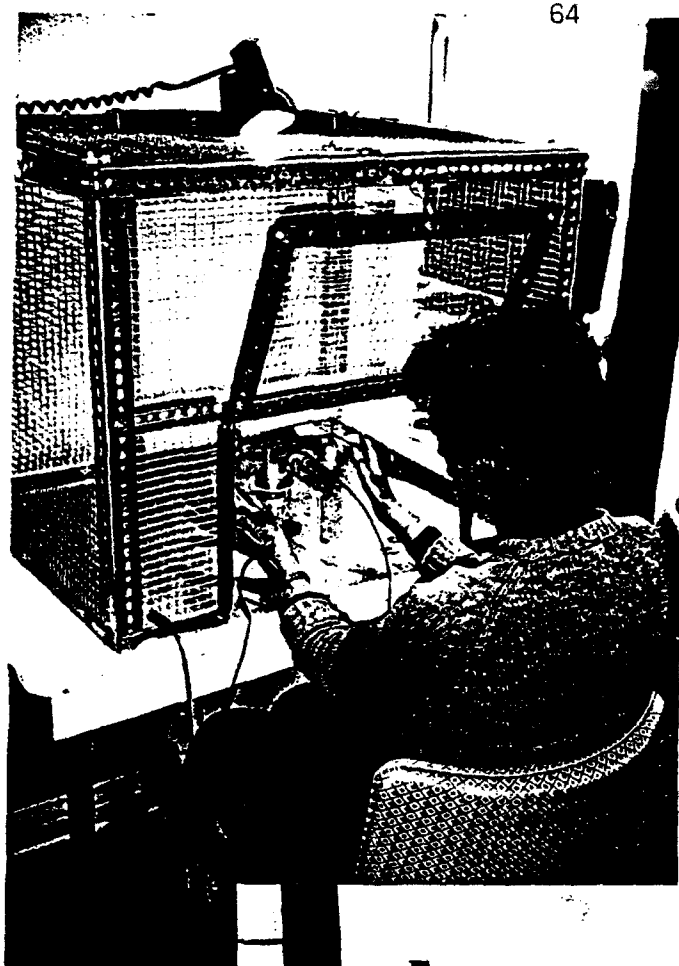


FIGURE 4. Map of the study areas in Iron and Dickinson Co. in Michigan's Upper Peninsula. Control sites: Site 1 = CL, Site 2 = C5. Experimental sites: Site 3 = F1, Site 4 = F2.



a



FIGURE 5a, b. Wire mesh Faraday cages, used to reduce exposure of nests to 60hz EM fields while nest architecture measurements are made in Crystal Falls.

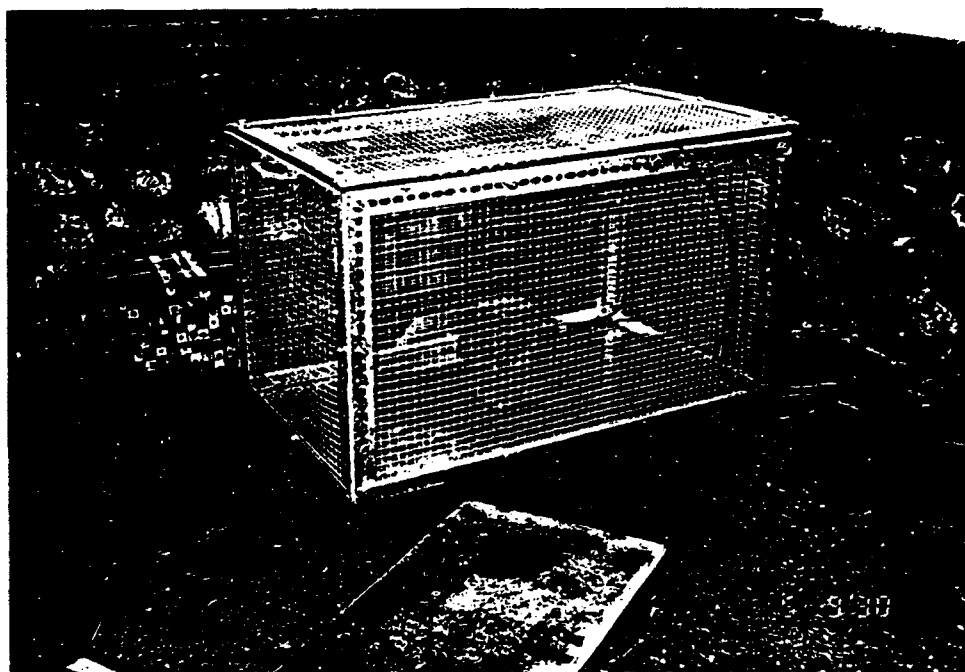


FIGURE 6. Wire mesh Faraday cage on front porch in Crystal Falls, used to store nests and cells just before and after nest measurements are made.

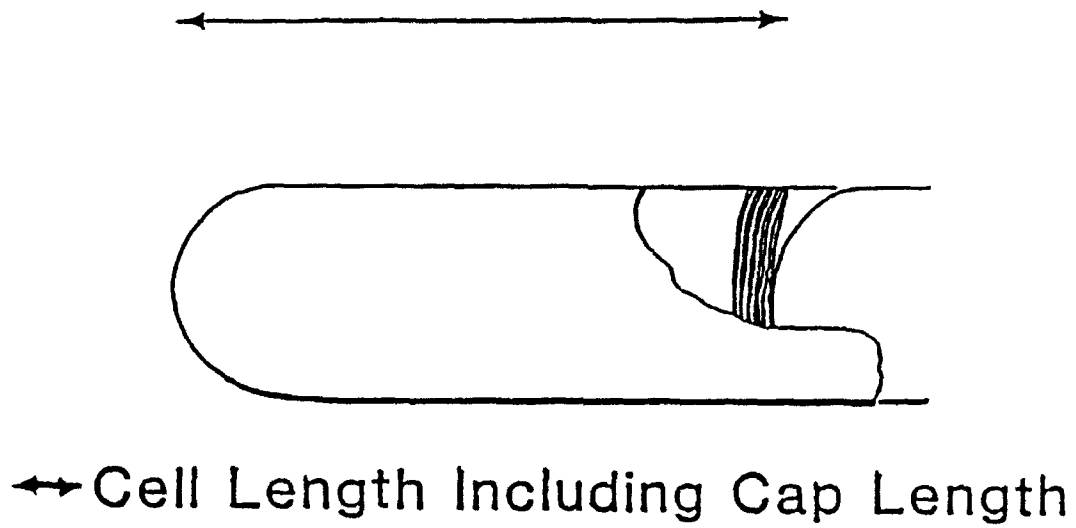


FIGURE 7. A single reproductive cell, indicating how cell lengths are measured.

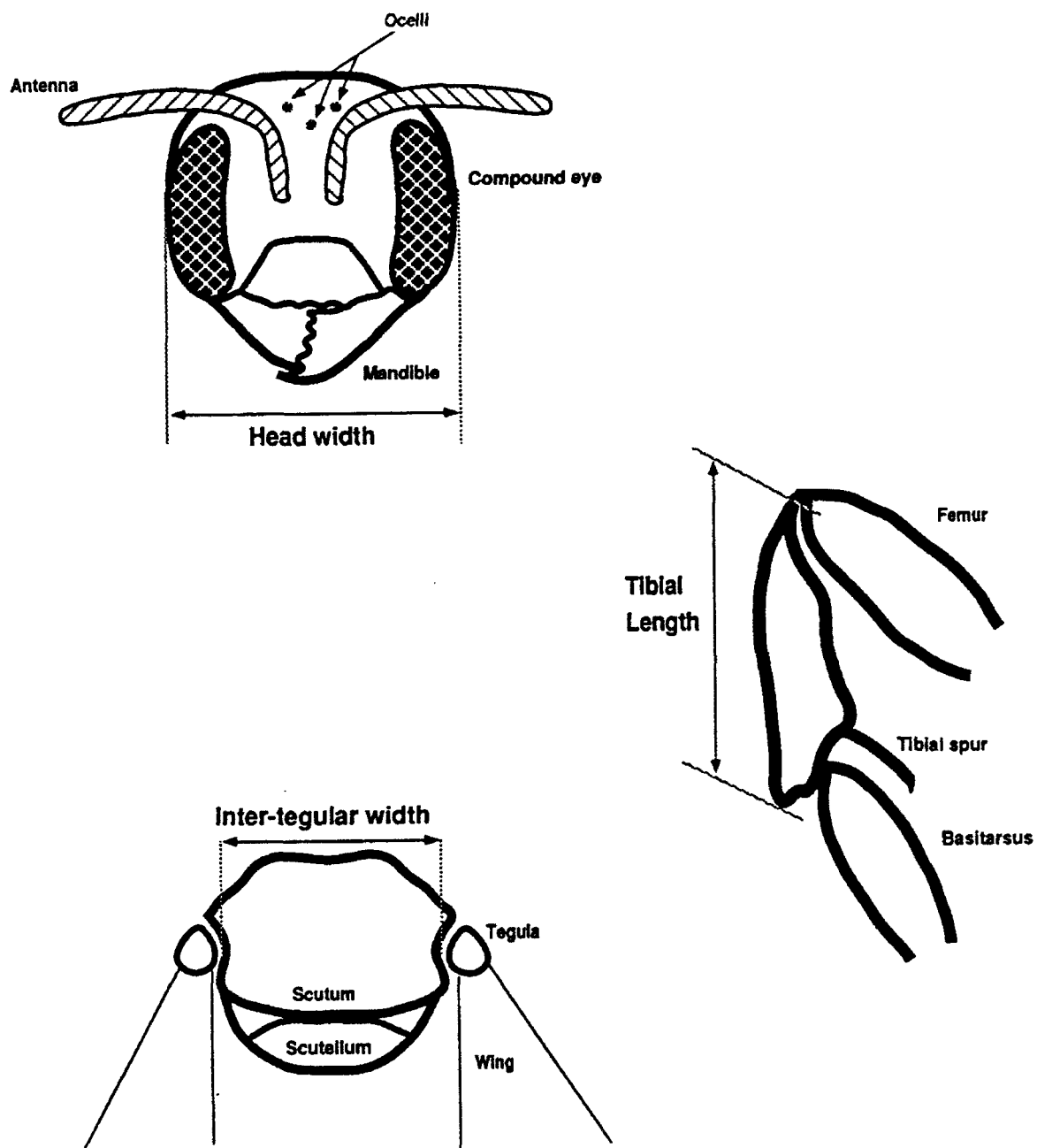


FIGURE 8. Hard body parts measured as indicators of body size.

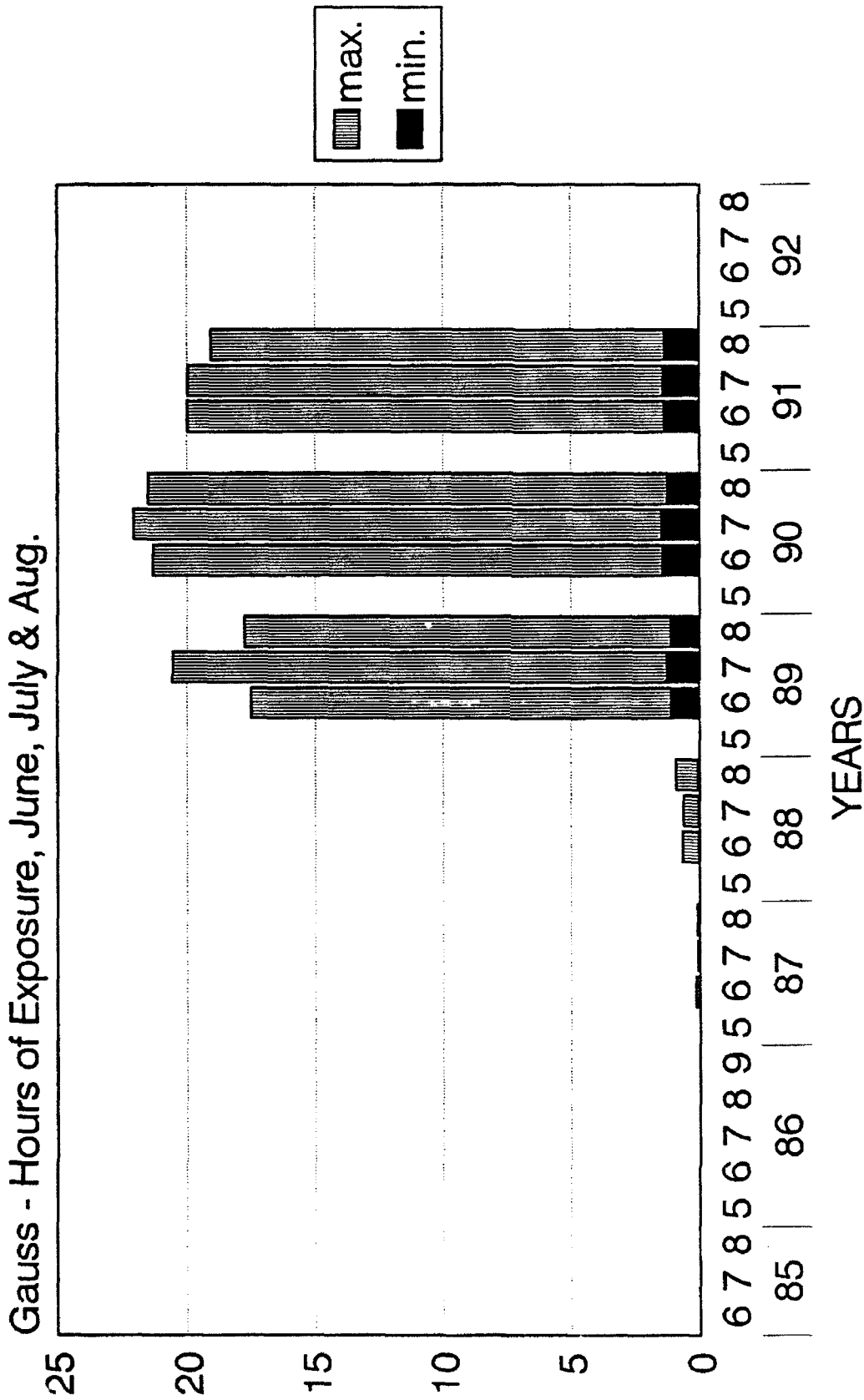


FIGURE 9. Cumulative magnetic field exposures (in Gauss-Hours) of foraging bees during the nesting season (June, July, August). A bee sitting directly under the antenna for the entire month would experience the maximum exposure plotted. A bee sitting on the hutch farthest from the antenna at the F2 site would experience the minimum exposure plotted. Most bees at the experimental sites would experience intermediate magnetic field exposure.

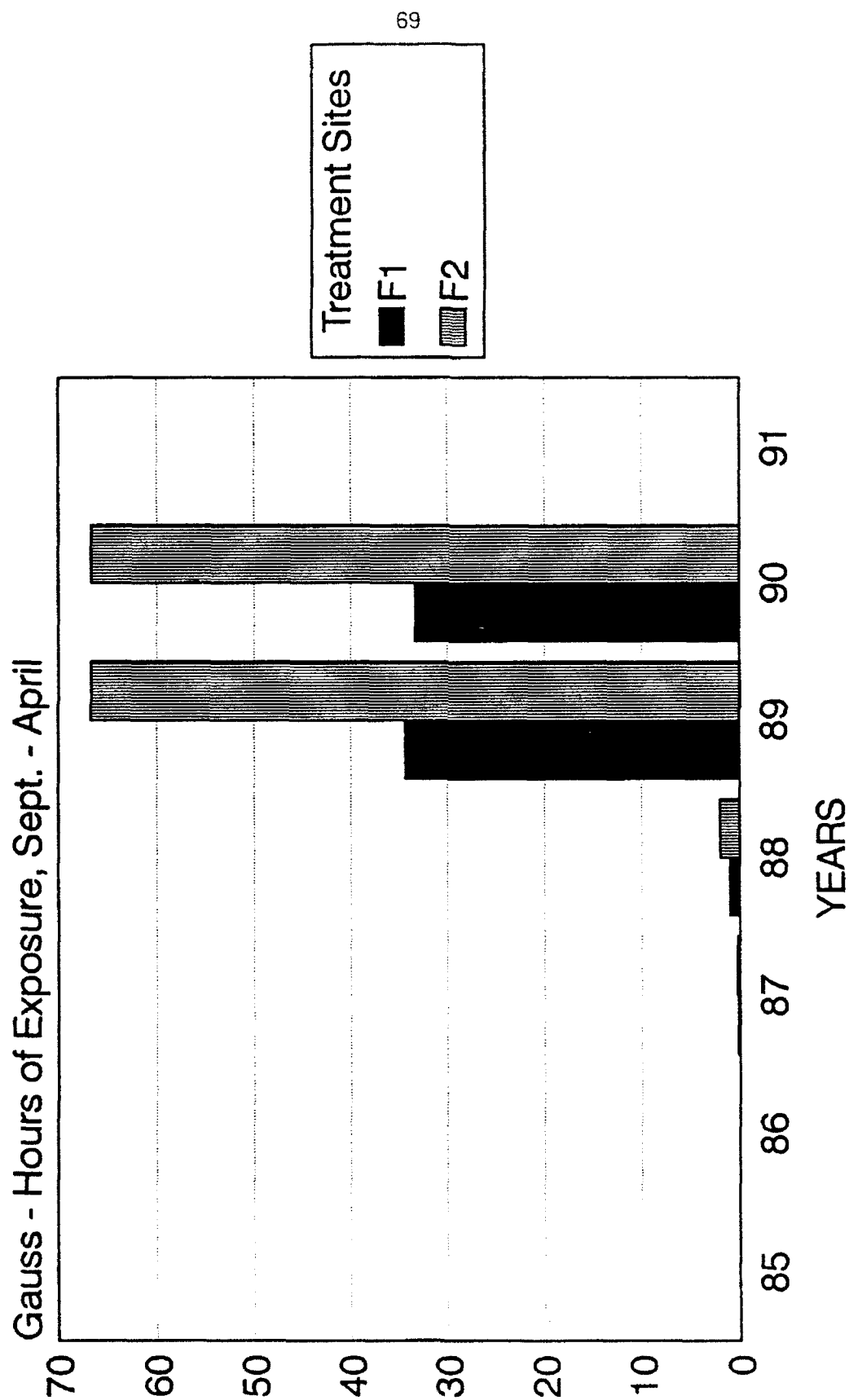


FIGURE 10. Cumulative magnetic field exposures (in Gauss-Hours) of overwintering prepupae between September and April.

TABLE 2: Basic form of the model used in GLM analyses. Degrees of freedom may differ for the two bee species. C represents constants calculated by SAS for each effect.

Source of Variation	Numerator df	Mixed Model F-Statistic
Exp (Fixed)	1	$MS \text{ Exp} / C*(MS \text{ Sites[Exp]} + C*(MS \text{ Error}))$
Sites[Exp] (Random)	2	$MS \text{ Sites[Exp]} / MS \text{ Error}$
Antenna (Fixed)	1	$MS \text{ Antenna} / C*(MS \text{ Yr[Antenna]} + C*(MS \text{ Measurer [Yr * Antenna]} + C*(MS \text{ Error})))$
Yr[Antenna] (Random)	3-6	$MS \text{ Yr[Antenna]} / C*(MS \text{ Measurer [Yr*antenna]} + C*(MS \text{ Error}))$
Measurer[Yr*Antenna] Error (Random - Cell length and plug length analyses only)	12-15	$MS \text{ Measurer[Yr*Antenna]} / MS \text{ Error}$
Exp * Antenna	1	$MS \text{ Exp * Antenna} / MS \text{ Error}$
<u>Other fixed effects, tested in some models:</u>		
Expected Sex Early vs. Late Season Complete vs. Incomplete nests	1	$MS \text{ Fixed Effect} / MS \text{ Error}$
<u>Other covariates, tested in some models:</u>		
Diameter Cells per Nest	1	$MS \text{ Covariate} / MS \text{ Error}$
<hr/> * $0.05 > P > 0.0011$ ** $0.001 > P > 0.00051$ *** $0.0005 > P > 0.0001$		

TABLE 3: Number of nests of M. relativa for which we have data on complete cell lengths, by site. (Number of hutches with 5 or more nests.)

Year	Control Sites		Test Sites	
	C5	CL	F1	F2
		<u>M. relativa</u>		
1983	--	27 (2)	128 (4)	17 (2)
1985	51 (5)	78 (6)	84 (5)	92 (6)
1986	49 (6)	51 (5)	42 (5)	80 (5)
1987	78 (5)	47 (5)	76 (4)	47 (5)
1988	85 (6)	59 (5)	83 (5)	51 (6)
1989	75 (6)	60 (5)	38 (3)	73 (6)
1990	70 (6)	82 (6)	54 (5)	123 (6)
1991	48 (4)	47 (6)	39 (5)	85 (6)
1992*	18 (2)	25 (2)	20 (1)	55 (5)

* Approximate numbers: Complete nests only; not yet measured.

TABLE 4: Number of nests of M. inermis for which we have data on complete cell lengths, by site. (Number of hutches with 5 or more nests.)

Year	Control Sites		Test Sites	
	C5	CL	F1	F2
		<u>M. inermis</u>		
1985 nests measured	23 (3)	17 (2)	160 (6)	88 (6)
nests constructed*	26	18	212	121
1986	15 (1)	2 (0)	40 (3)	65 (4)
1987	56 (3)	25 (3)	122 (5)	108 (6)
1988	30 (3)	7 (0)	54 (2)	127 (5)
1989	106 (6)	23 (3)	172 (6)	262 (6)
1990	163 (6)	51 (3)	237 (6)	382 (6)
1991	138 (5)	54 (5)	187 (6)	374 (6)
1992**	35 (4)	20 (2)	104 (6)	152 (6)

* Some 1985 nests were not measured because they were used in a study of diapause. I do not have these nests, nor do I have the data from the diapause study.

** Approximate numbers: Complete nests only; not yet measured.

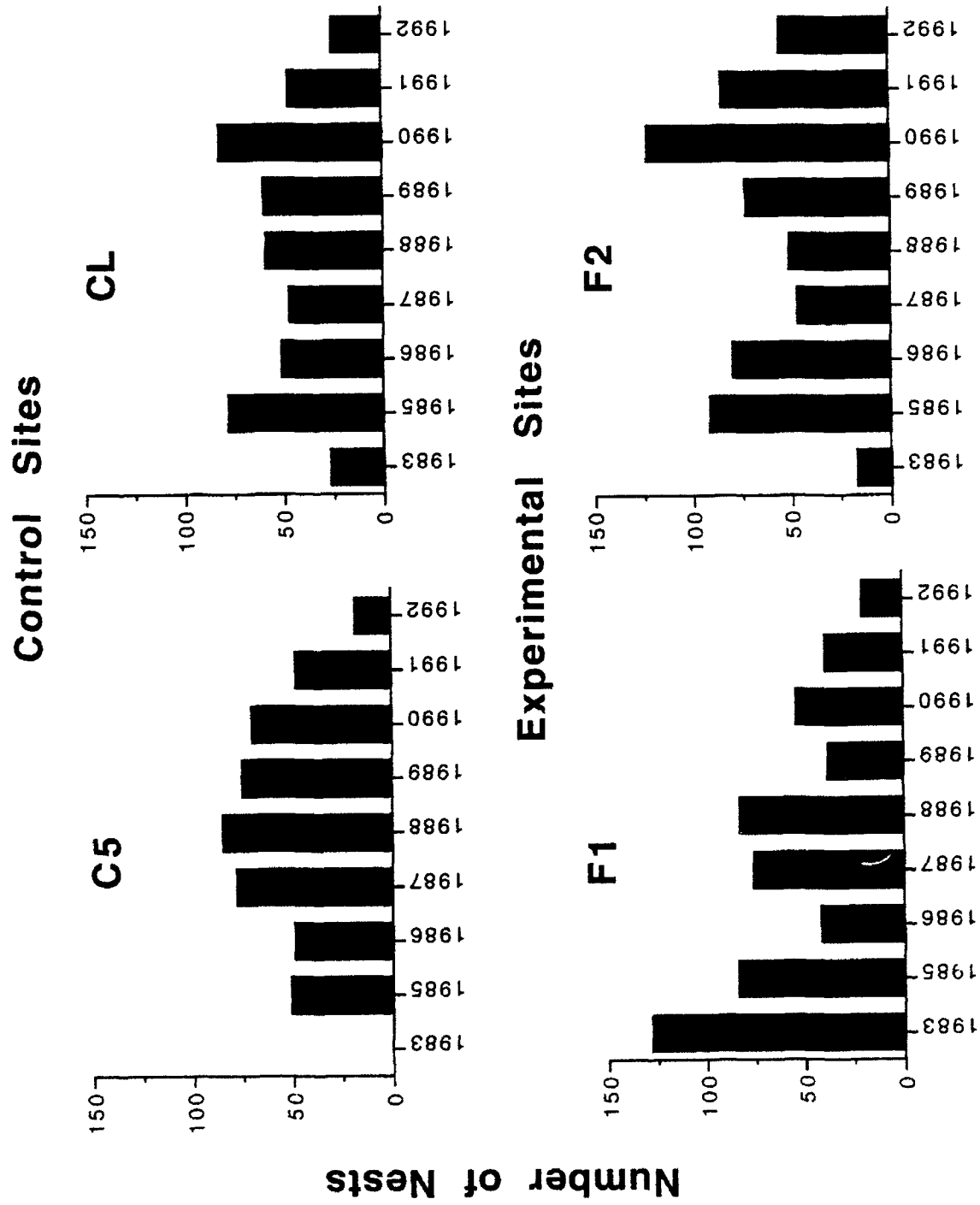


FIGURE 11. Number of nests of *M. relativa* constructed at four sites, 1983-1992.

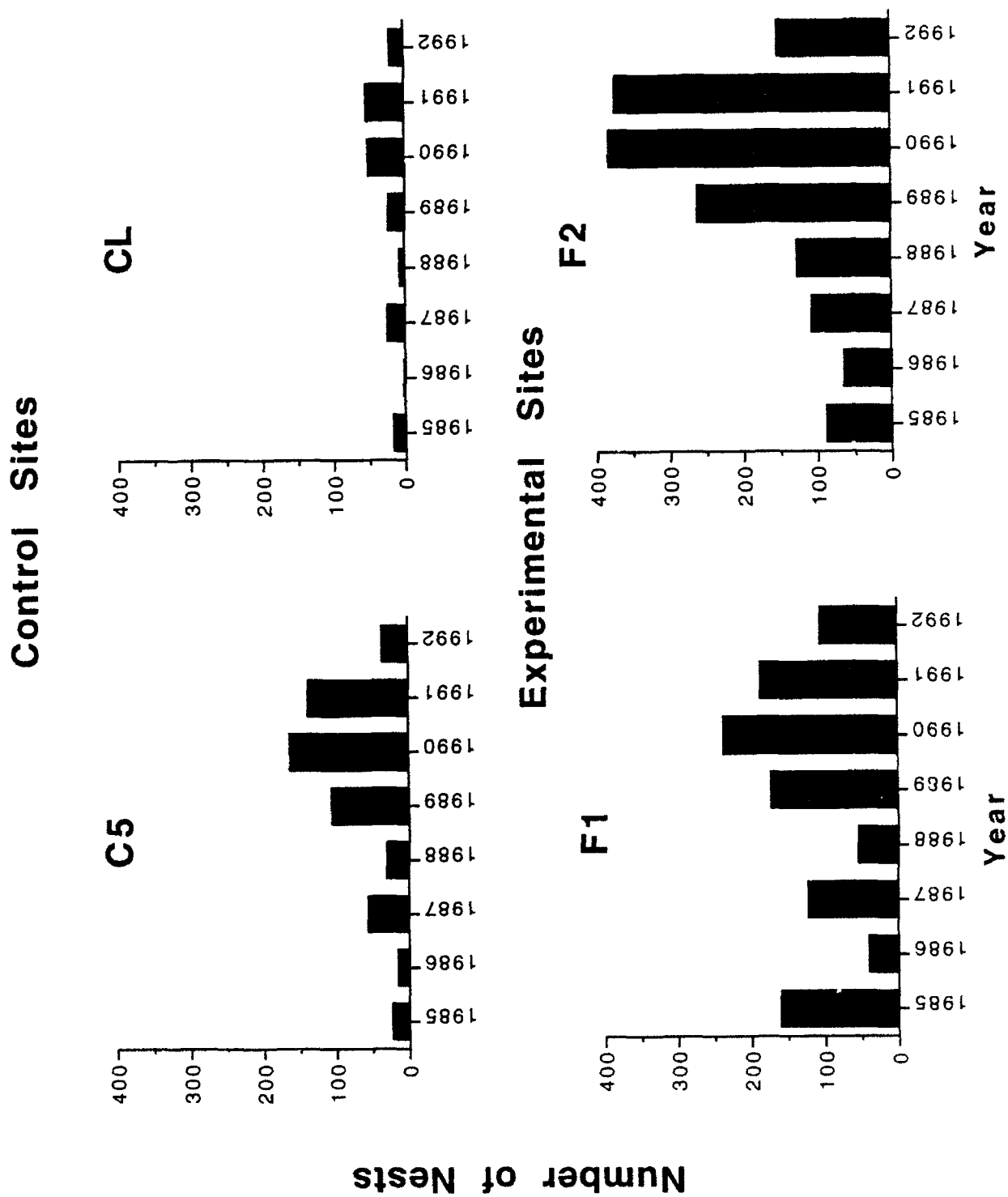


FIGURE 12. Number of nests of *M. inermis* constructed at four sites, 1985-1992.

CUMULATIVE PRECIPITATION

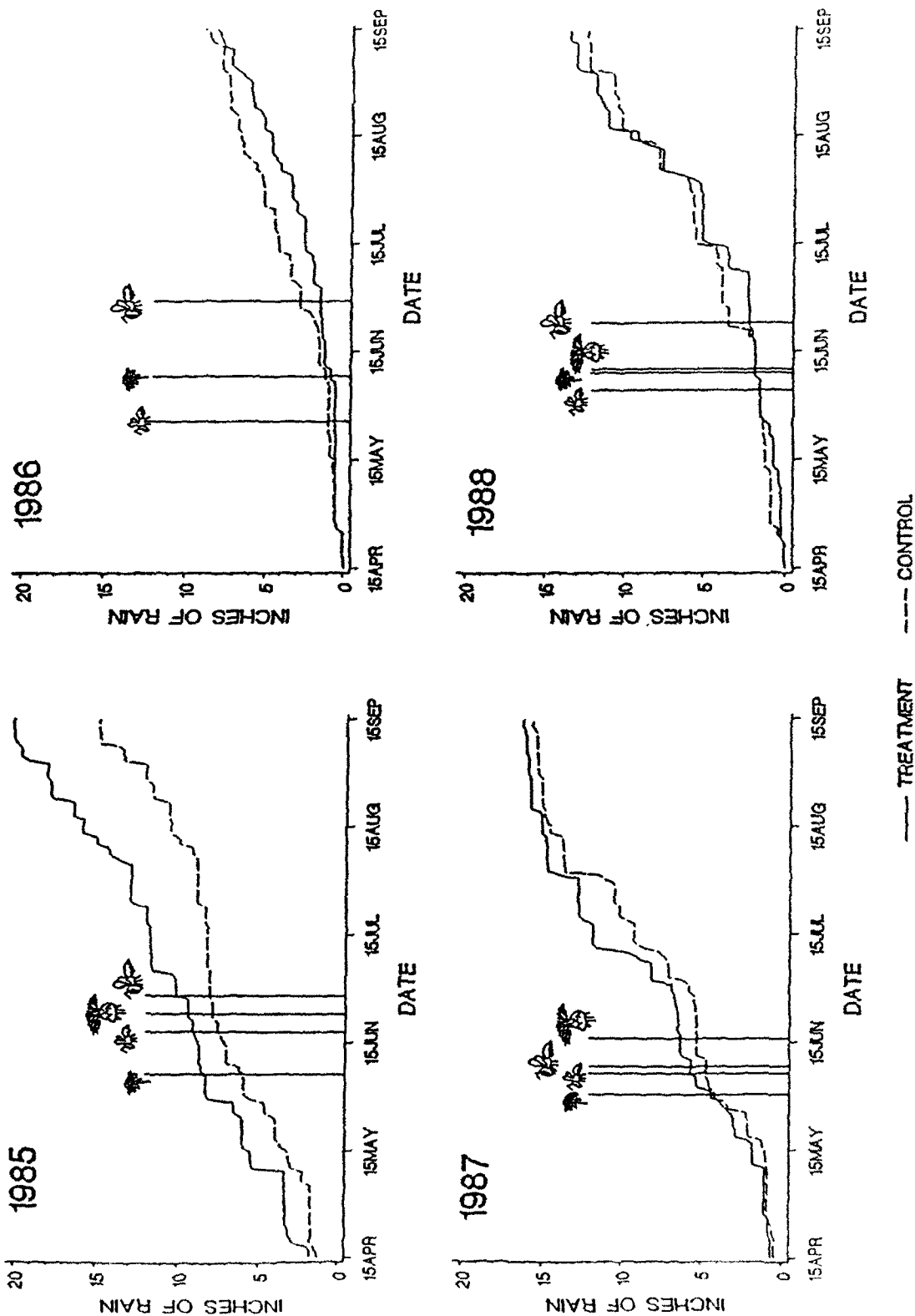


FIGURE 13. Cumulative precipitation at MTU pine plantations. Vertical lines indicate date of first nest of *M. relativa* (small bee) and *M. inermis* (large bee), and first bloom of orange hawkweed (*Hieracium aurantiacum*) and thistle (*Cirsium palustre*).

CUMULATIVE PRECIPITATION

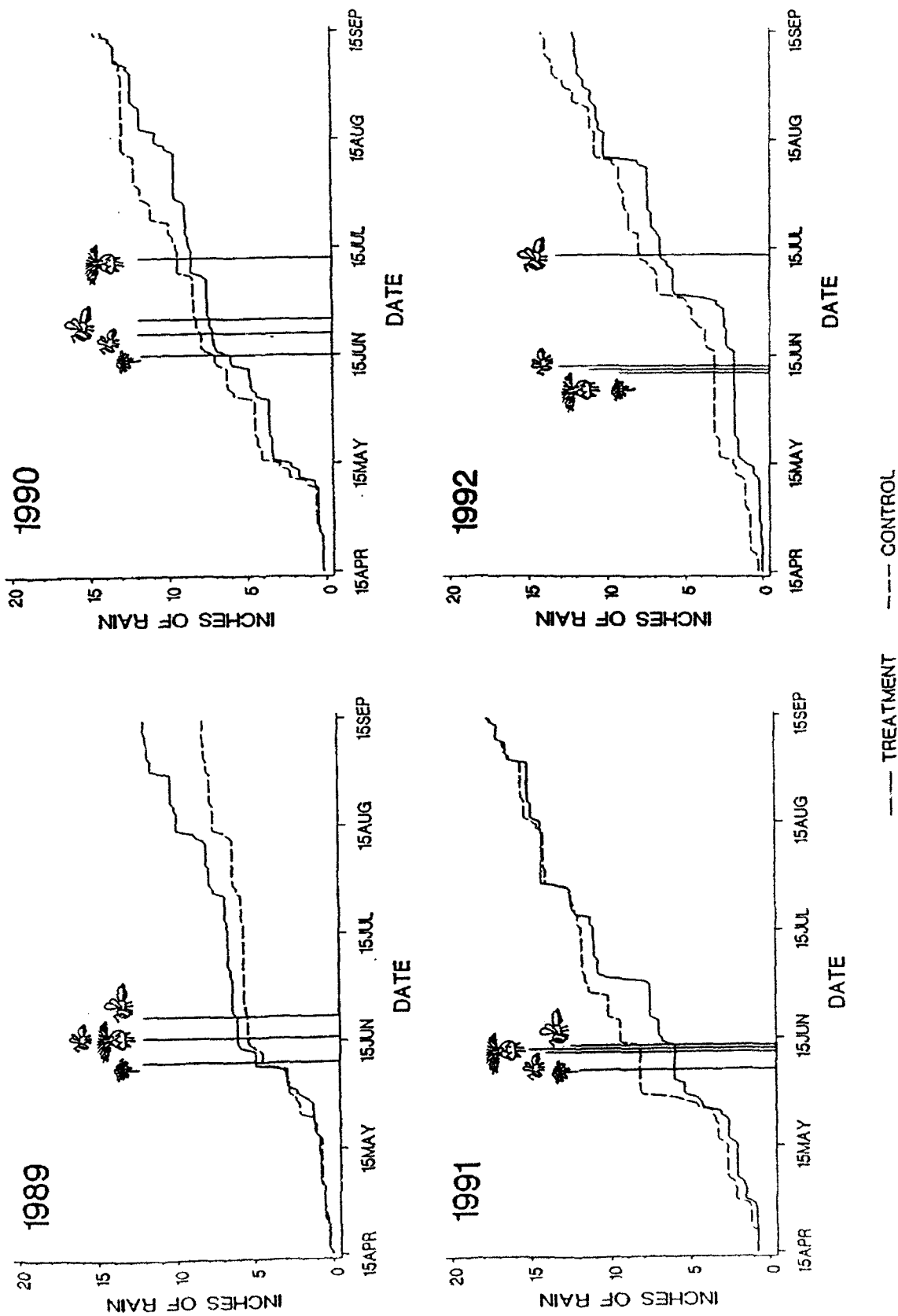


FIGURE 13. (cont.). Cumulative precipitation at MTU pine plantations.

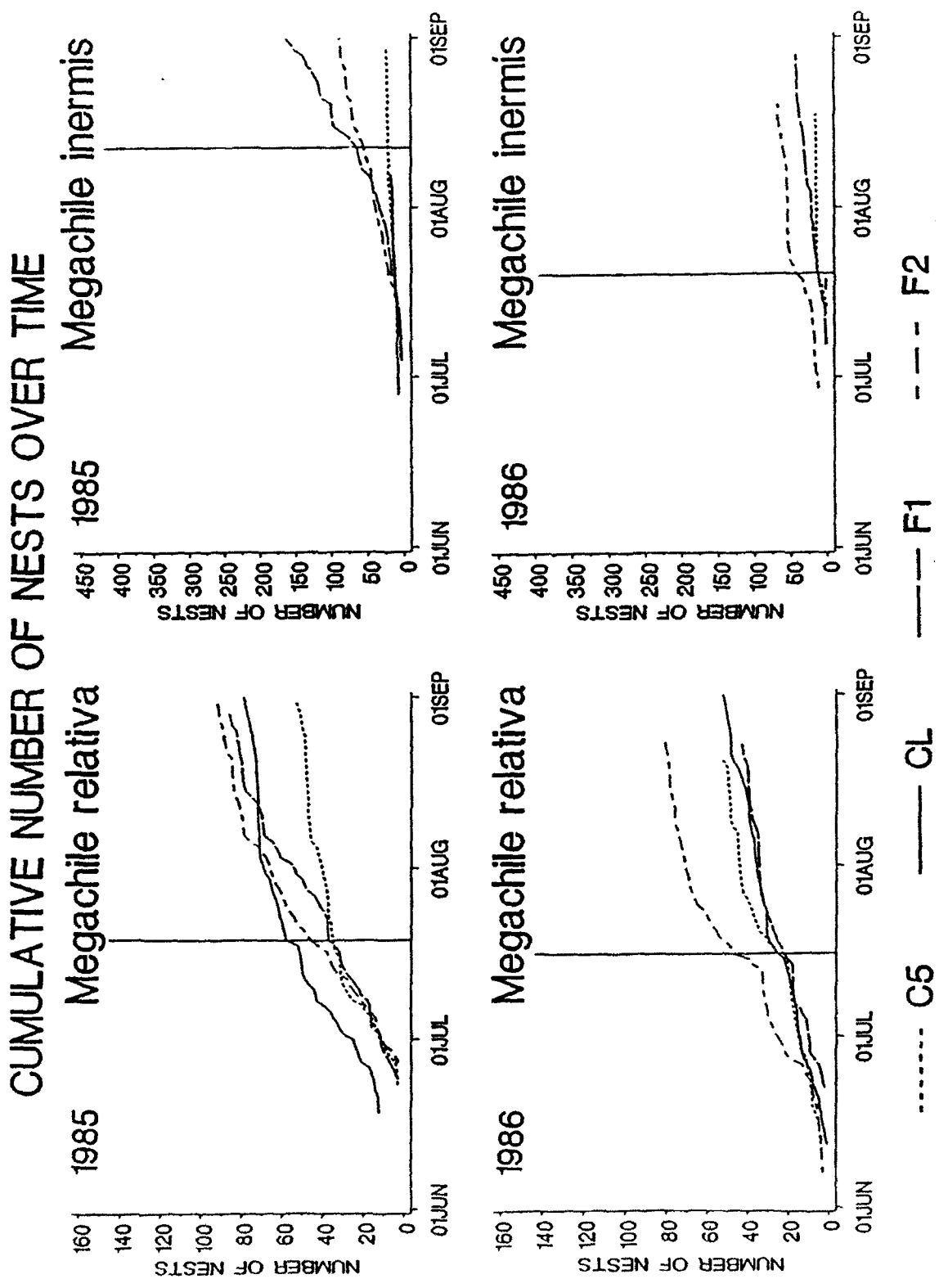


FIGURE 14a. Cumulative number of nests of *M. relativa* and *M. inermis* at each site, 1985-1986. Note different scales for each species. Vertical lines indicate date on which the last early season nests were begun for each site.

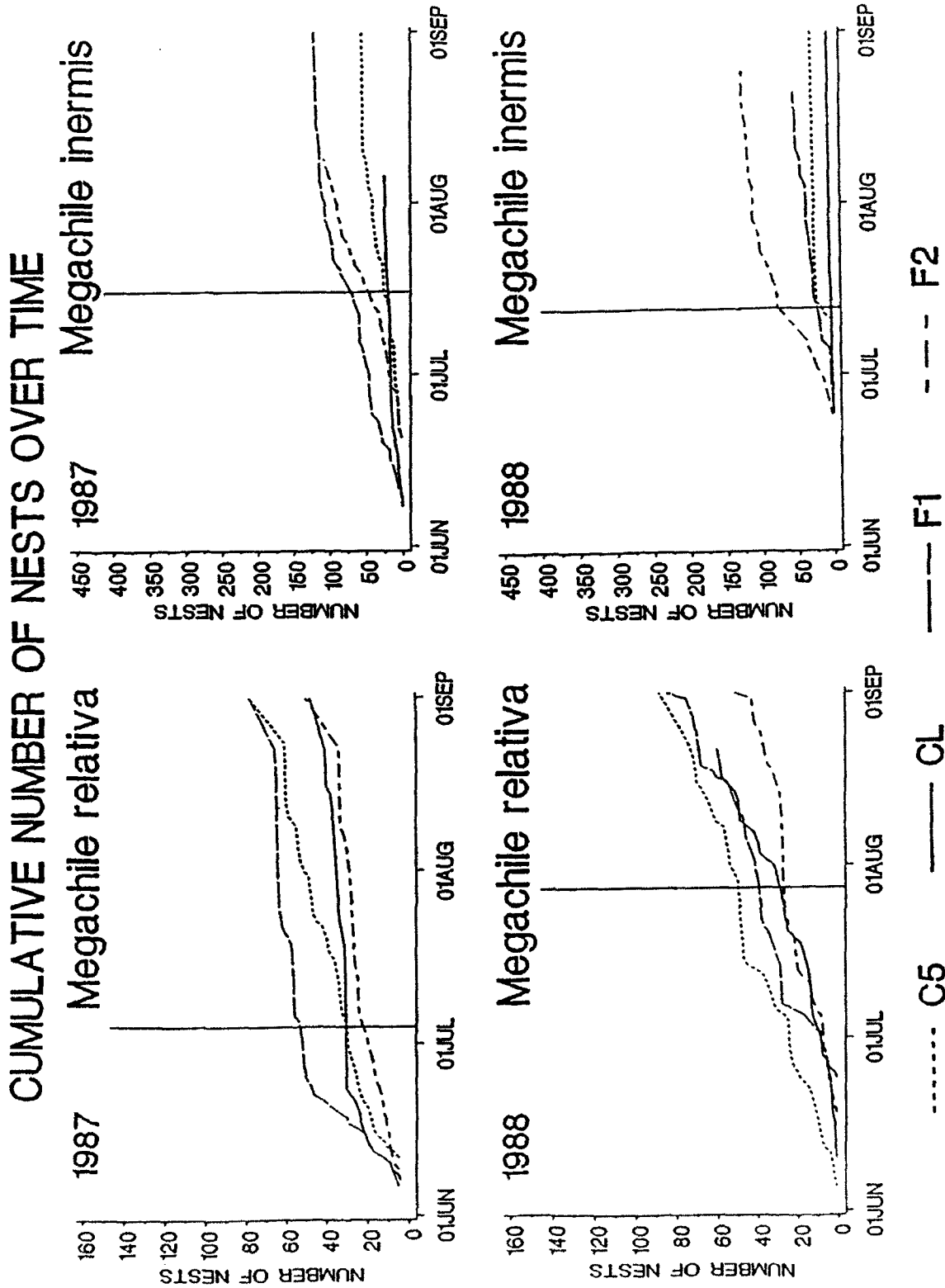


FIGURE 14b. Cumulative number of nests of *M. relativa* and *M. inermis* at each site, 1987-1988. Note different scales for each species. Vertical lines indicate date on which the last early season nests were begun for each site.

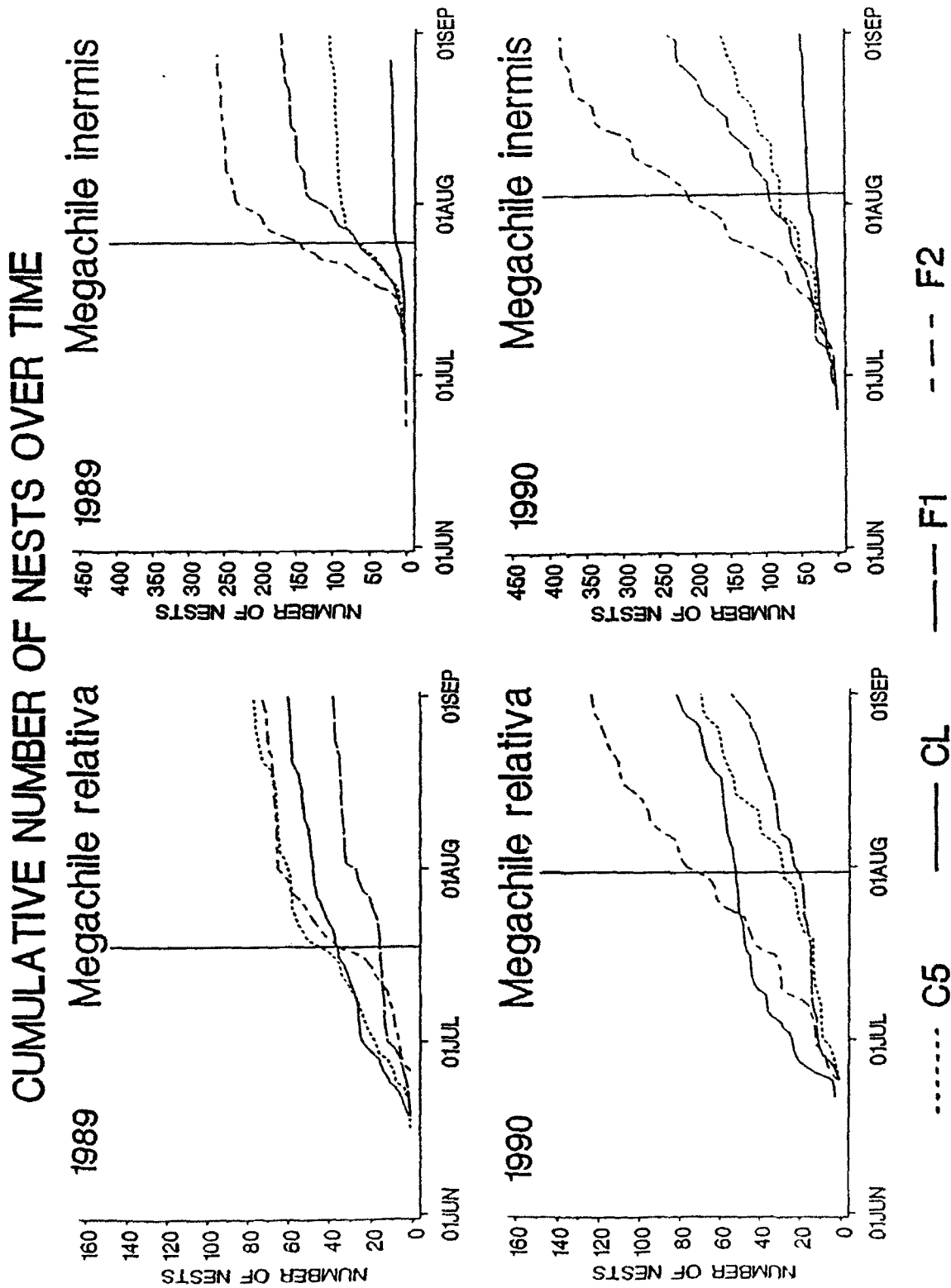


FIGURE 14c. Cumulative number of nests of *M. relativa* and *M. inermis* at each site, 1989-1990. Note different scales for each species. Vertical lines indicate date on which the last early season nests were begun for each site.

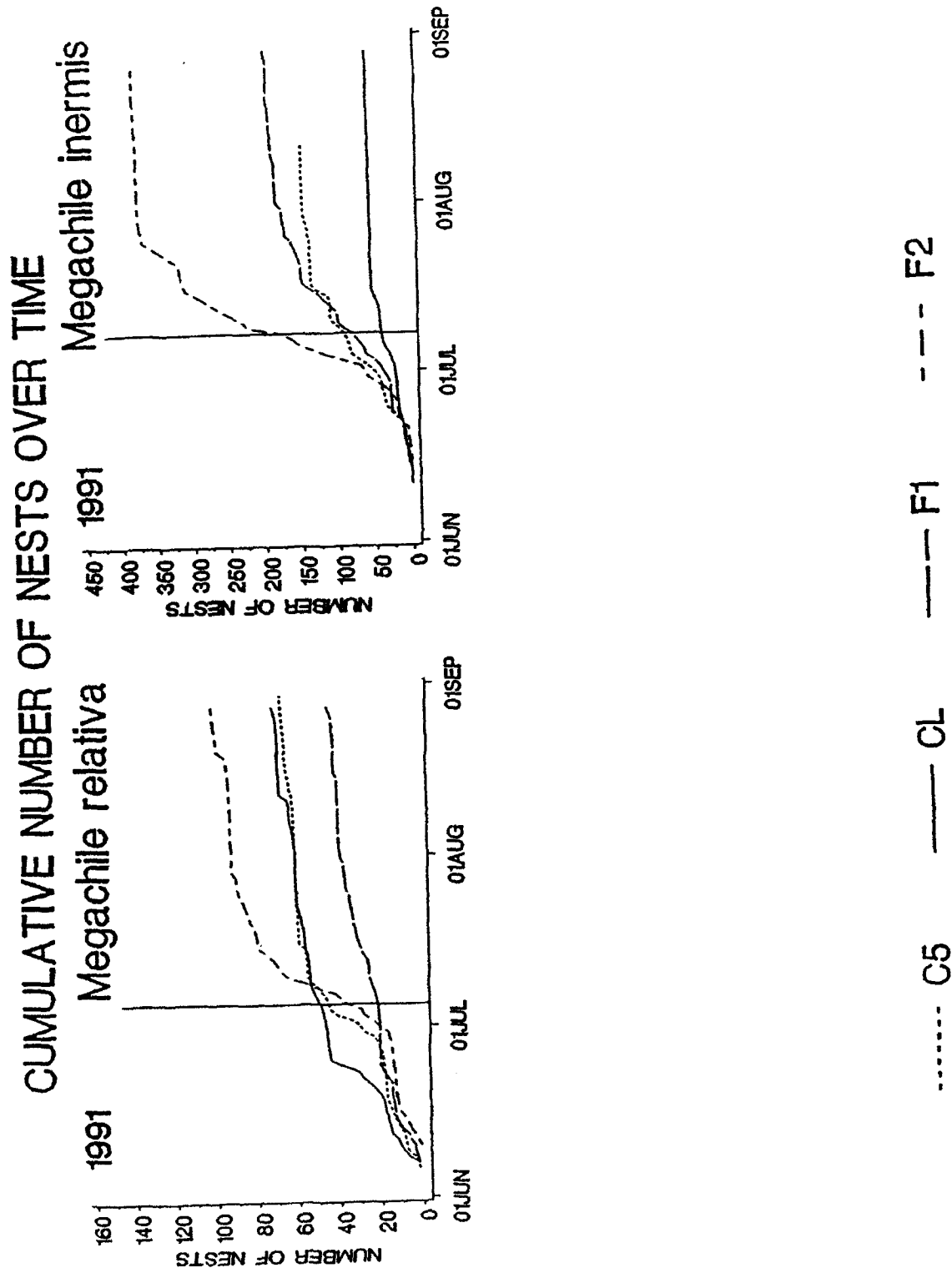


FIGURE 14d. Cumulative number of nests of *M. relativa* and *M. inermis* at each site, 1991. Note different scales for each species. Vertical lines indicate date on which the last early season nests were begun for each site.

Mean Cell Lengths *Megachile relativa*

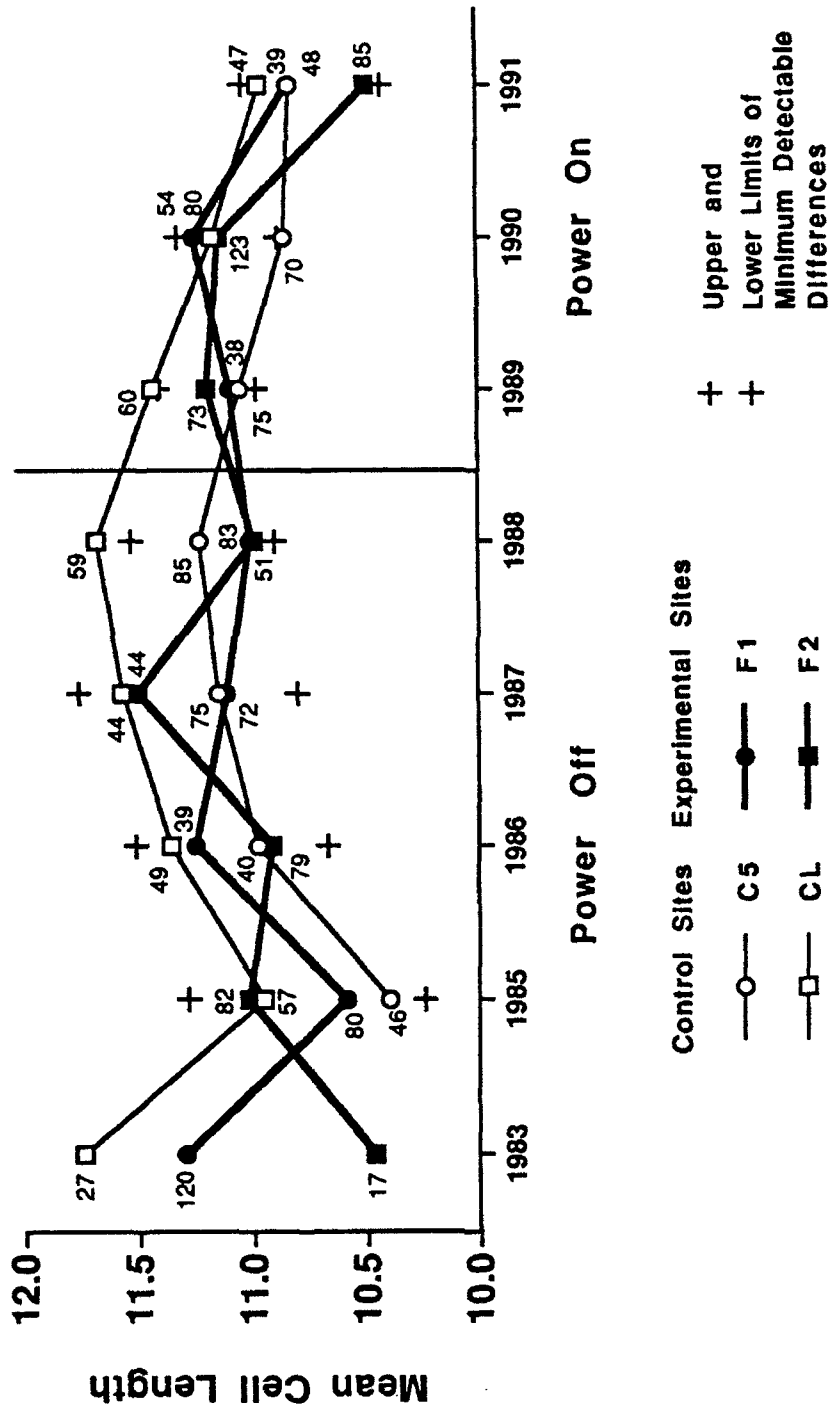


FIGURE 15. Mean cell length for *M. relativa* nests, 1983-1991, all cells. Numbers indicate sample sizes; + indicates the upper and lower limits to the minimum detectable difference between experimental and control sites for each year.

TABLE 5: GLM of mean cell length for all cells from 1983-1991 *M. relativa* nests.

CELL LENGTHS				
Source of variation	Numerator df	MS	F	P>F
Exp	1	0.38	0.05	0.8464
Site [Exp]	2	7.80	10.79	0.0001***
Antenna	1	2.37	0.75	0.4088
Year [Antenna]	6	9.36	4.21	0.0078*
Measurer [Year * Antenna]	17	2.40	3.33	0.0001***
Exp * Antenna	1	3.67	5.08	0.0244*
Diameter	1	16.26	22.51	0.0001***
Complete vs. incomplete	1	5.62	7.78	0.0053*
Cells per nest	1	58.29	80.70	0.0001***
Early vs. Late Season	1	52.62	72.85	0.0001***
Model	32	268.45	11.61	0.0001***
Error	1908	1378.10		
$\bar{X} = 11.6$ mm	CV=7.7	$r^2=0.16$	For $\alpha=0.05$ Power of Exp * Antenna= 0.61	

TABLE 5 (continued)

Parameter	Estimates:	T for H ₀ : Parameter = 0	PR > T	\bar{X}	SD
Exp * Antenna					
Control Off	0.099	1.00	0.3184	11.21	0.92
Control On	0.00	--	--	11.06	0.89
Experimental Off	0.00	--	--	11.04	0.95
Experimental On	0.00	--	--	11.00	0.89
Year (Antenna)					
Off: 1983	0.508	0.59	0.5524	11.28	1.11
1985	-0.467	-4.06	0.0001***	10.76	0.96
1986	-0.092	-0.57	0.5680	11.09	0.81
1987	0.198	1.77	0.0776	11.28	0.82
1988	0.0	--	--	11.22	0.92
On: 1989	0.243	1.90	0.0572	11.20	0.83
1990	0.079	0.71	0.4758	11.10	0.90
1991	0.0	--	--	10.73	0.87
Site					
C5	0.0	--	--	11.32	0.94
CL	0.248	4.16	0.0001***	10.97	0.84
F1	0.111	1.96	0.0498*	11.06	0.92
F2	0.0	--	--	10.99	0.93
Diameter	-0.170	-4.74	0.0001***		
Complete vs. Incomplete	0.130 0.0	2.79 --	0.0053** --		
Cells per nest	-0.082	-8.98	0.0001***		
Early Season vs. Late Season	0.349 0.0	8.54 --	0.0001*** --		

TABLE 6: GLM of mean cell length for all cells from 1983-1991 *M. relativa* nests; expected sex included in model.

CELL LENGTHS				
Source of variation	Numerator df	MS	F	P>F
Exp	1	5.38	0.78	0.4679
Site [Exp]	2	8.62	13.69	0.0001***
Antenna	1	4.43	0.71	0.4343
Year [Antenna]	6	7.90	2.84	0.0485*
Measurer [Year * Antenna]	15	2.63	4.17	0.0001***
Exp * Antenna	1	4.09	6.50	0.0109*
Sex	1	119.25	189.35	0.0001***
Exp * Sex	1	0.60	0.95	0.3304
Antenna * Sex	1	2.20	3.45	0.0619
Exp * Antenna * Sex	1	1.63	2.59	0.1080
Diameter	1	24.52	38.94	0.0001***
Complete vs. incomplete	1	3.88	6.17	0.0131*
Cells per nest	1	37.11	58.93	0.0001***
Early vs. Late Season	1	37.33	59.27	0.0001***
Model	34	10.89	17.30	0.0001***
Error	1582	0.63		
$\bar{X} = 11.1$ mm	CV=7.2	$r^2=0.27$	For $\alpha=0.05$ Power of Exp * Antenna=0.72	

TABLE 6 (continued)

Parameter	Estimate:	T for H ₀ :		\bar{X}	SD
		Parameter = 0	PR> T		
Exp * Antenna					
Control Off	0.099	1.00	0.3184	11.24	0.96
Control On	0.0	--	--	11.10	0.87
Experimental Off	0.0	--	--	11.02	0.96
Experimental On	0.0	--	--	10.96	0.83
Site [Exp]:					
C5	0.0	--	--	11.04	0.89
CL	0.30	4.62	0.0001***	11.38	0.93
F1	0.13	2.28	0.0227*	11.06	0.94
F2	0.0	--	--	10.93	0.88
Sex: Female	0.60	6.06	0.0001***	11.55	0.90
Male	0.00	--	--	10.90	0.87
Diameter	-0.22	-6.24	0.0001***		
Cells per nest	-0.07	-7.68	0.0001***		
Early Season vs.	0.33	7.70	0.0001***		
Late Season	0.0	--	--		

TABLE 7: Differences between measurers in mean cell lengths for M. relativa.

Measurer	Mean Cell Lengths mm	SD.	No. Nests Measured
JH (1983)	11.1	—	1
MA (1983)	11.3	1.1	162
VS (1983)	11.7	—	1
ER (1985)	10.8	1.0	85
ND (1985)	10.6	1.0	99
KS (1985)	10.9	0.8	81
JZ (1986)	11.3	0.7	64
KS (1986)	11.2	0.9	58
LS (1986)	10.7	0.8	49
MS (1986)	11.1	0.7	36
KS (1987)	11.3	0.9	99
LS (1987)	11.2	0.7	28
VS (1987)	11.3	0.8	108
BZ (1988)	11.0	1.0	68
KS (1988)	11.4	0.9	80
VS (1988)	11.2	0.8	130
BZ (1989)	11.0	0.8	79
KS (1989)	11.4	0.8	83
VS (1989)	11.1	0.8	84
JR (1990)	11.4	1.0	102
KS (1990)	10.9	0.8	72
VS (1990)	11.0	0.9	153
BZ (1991)	10.5	0.8	69
KS (1991)	10.7	1.0	55
VS (1991)	10.9	0.8	95

TABLE 8: Two-Way, Model II ANOVA partitioning the variance in cell length within- and between-measurer.

CELL LENGTHS				
Source of Variance	Numerator df	MS	F	P>F
Between Measurers	3	9.587	65.39	0.0001***
Between Cells	38	7.540	51.42	0.0000***
Within Measurer (Error)	355	0.147		
	CV = 3.6	$r^2 = 0.86$		
Between Measurers	$s^2 + 2.55s_{mc}^2 + 39(2.55)s_m^2$		0.095	9.8%
Between Cells	$s^2 + 2.55s_{mc}^2 + 4(2.55)s_c^2$		0.725	75.0%
Within Measurer (Error)	$s^2 + 2.55s_{mc}^2$		0.147	15.2%

Mean Cell Lengths *Megachile inermis*

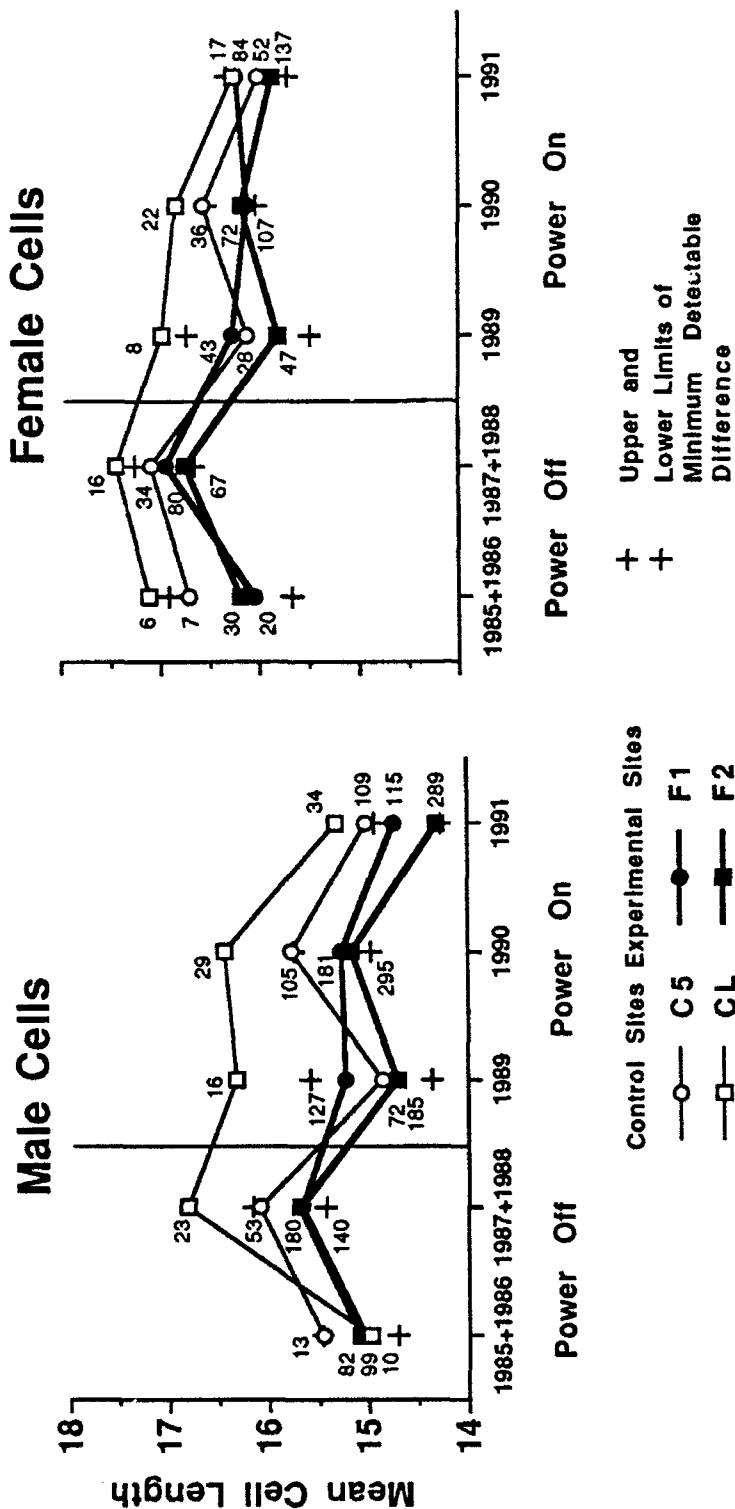


FIGURE 16. Mean cell length for *M. inermis* nests by sex; diameters >9.5mm. Numbers indicate sample sizes; + indicate upper and lower limits to the minimum detectable difference between experimental and control sites for each year.

TABLE 9: GLM of mean cell lengths for M. inermis nests; diameters > 9.5mm; expected sex included in the model.

CELL LENGTHS

Source of variation	Numerator df	MS	F	P>F
Exp	1	52.02	4.01	0.1731
Site [Exp]	2	27.40	28.32	0.0001***
Antenna	1	51.33	2.25	0.2468
Year [Antenna] 85+86, 87+88, 89, 90, 91	3	59.14	1.11	0.3823
Measurer [Year * Antenna]	12	57.85	59.79	0.0001***
Exp. * Antenna	1	0.37	0.39	0.5344
Sex	1	409.57	423.28	0.0001***
Exp. * Sex	1	3.70	3.82	0.0507
Antenna * Sex	1	0.06	0.06	0.8025
Exp * Antenna * Sex	1	0.06	0.07	0.7969
Diameter	1	6.80	7.03	0.0081*
Complete vs. incomplete	1	30.95	31.99	0.0001***
Cells per nest	1	277.95	287.26	0.0001***
Early vs. Late Season	1	5.08	5.25	0.0221*
Model	28	88.81	91.78	0.0***
Error	3041	0.97		
$\bar{X} = 15.5\text{mm}$	CV = 6.3	$r^2 = 0.46$		

TABLE 9 (continued)

Parameter	Estimate	T for H_0 : Parameter = 0	PR > T	\bar{X}	SD
Site [Exp]:					
C5	0.0	—	—	15.71	1.30
CL	0.475	5.51	0.0001***	16.35	1.35
F1	0.214	5.09	0.0001***	15.58	1.26
F2	0.0	—	—	15.26	1.32
Sex					
F	1.21	22.55	0.0001***	16.32	1.18
M	0.0	—	—	15.15	1.23
Diameter	0.11	2.65	0.0081*		
Complete vs. incomplete					
	0.380	5.66	0.0001***		
	0.0	—	—		
Season:					
Early vs	0.097	2.29	0.0221*		
Late	0.0	—	—		

TABLE 10: Differences between observers in mean male cell lengths for M. inermis, bore diameters >9.5mm.

Measurer	Mean Cell Lengths m m	S.D.	No. Nests Measured
LS (1985-86)	15.0	1.1	138
MS (1985-86)	15.4	1.3	70
JZ (1985-86)	16.1	1.3	28
KS (1985-86)	16.5	1.2	31
LS (1987-88)	15.5	1.1	53
VS (1987-88)	15.8	1.1	286
BZ (1987-88)	15.8	1.1	34
KS (1987-88)	16.9	1.2	220
VS (1989)	15.1	1.1	260
BZ (1989)	14.7	1.3	137
KS (1989)	16.1	1.2	129
JR (1990)	15.7	1.1	284
KS (1990)	15.9	1.2	258
VS (1990)	15.3	1.1	305
BZ (1991)	14.4	1.3	311
KS (1991)	15.8	1.2	237
VS (1991)	15.2	1.1	289

Table 11: R^2 for regressions of body lengths on weight.

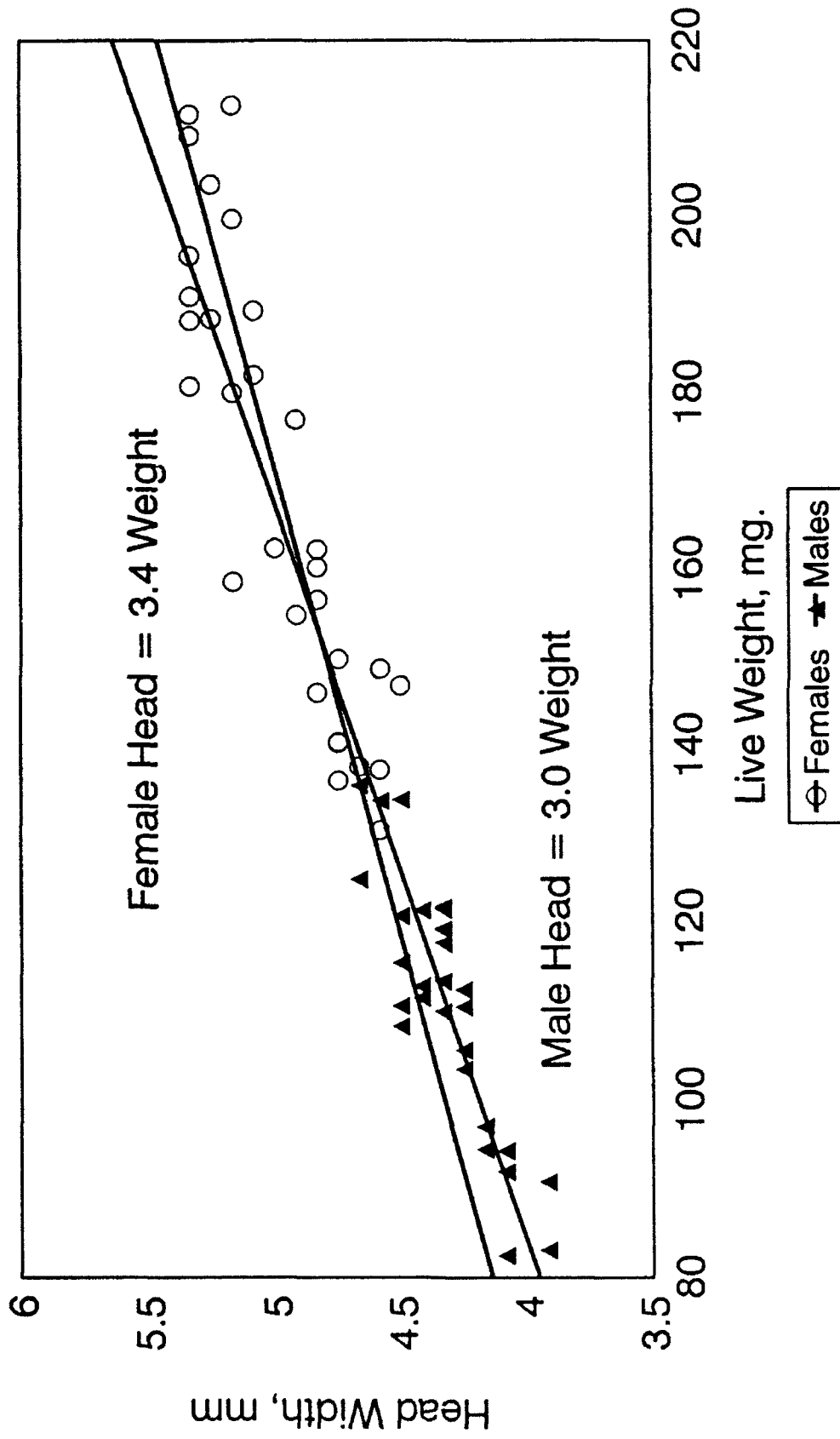
Species	<i>Megachile inermis</i>		<i>Megachile relativa</i>	
Sex	♀	♂	♀	♂
Head vs. Dry Wt.	0.55	0.73	0.71	0.92
Head vs. Live Wt.	0.77	0.76	0.77	0.93
Thorax vs. Dry Wt.	0.43	0.48	0.78	0.88
Thorax vs. Live Wt.	0.66	0.51	0.82	0.89
Tibia vs. Dry Wt.	0.41	0.66	0.62	0.85
Tibia vs. Live Wt.	0.48	0.80	0.70	0.85

Table 12: R^2 for regressions between body lengths, and between dry and live weights.

Species	<i>Megachile inermis</i>		<i>Megachile relativa</i>	
Sex	♀	♂	♀	♂
Head vs. Thorax	0.74	0.48	0.82	0.90
Head vs. Tibia	0.54	0.72	0.78	0.89
Thorax vs. Tibia	0.60	0.48	0.68	0.82
Dry Wt. vs. Live Wt.	0.75	0.89	0.95	0.98

Table 13: Ratio of highest to lowest values of weight and lengths.

Species	<i>Megachile inermis</i>		<i>Megachile relativa</i>	
Sex	♀	♂	♀	♂
Live Weight	1.63	1.64	2.28	2.48
Head	1.19	1.19	1.25	1.32
Thorax	1.21	1.18	1.26	1.35
Tibia	1.32	1.19	1.27	1.38

FIGURE 17. Regression of head length on live weight for *M. inermis*

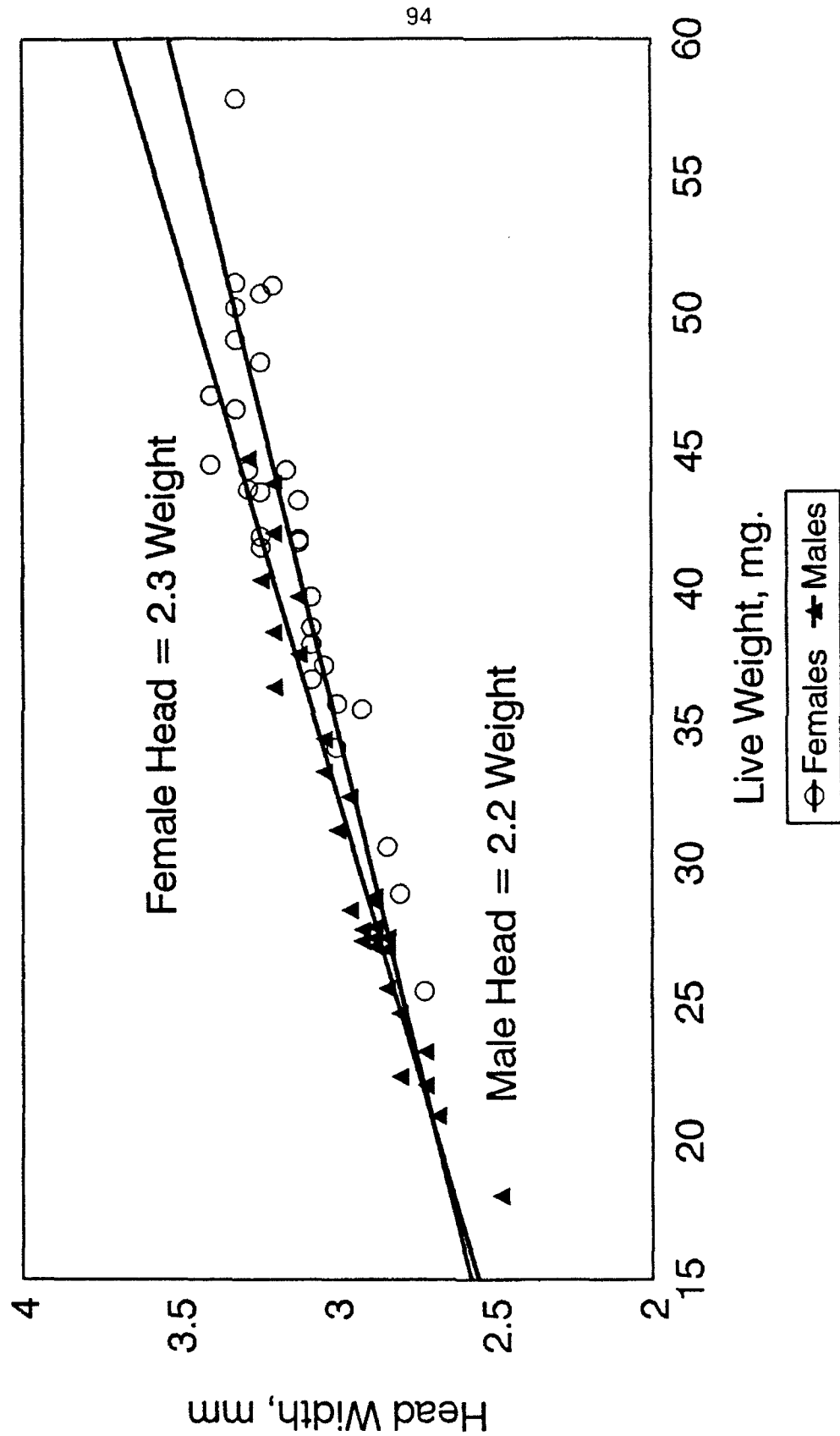


FIGURE 18. Regression of head length on live weight for *M. relativa*.

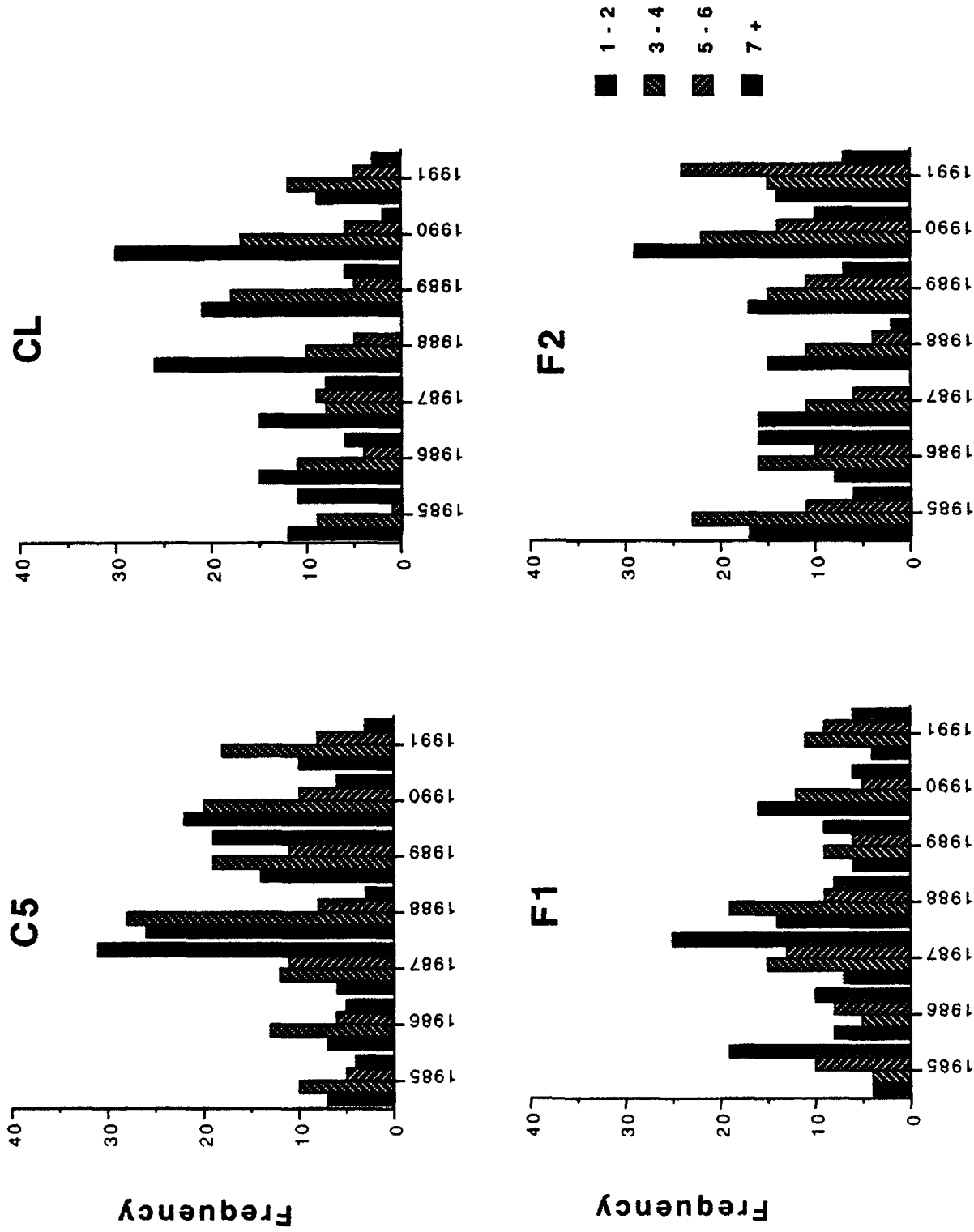
FIGURE 19. Number of complete nests of *M. relativa* with 1-2, 3-4, 5-6 or 7+ cells.

TABLE 14: Categorical modeling of number of cells per complete nest of Megachile relativa, 1985-1991.

NUMBER OF CELLS PER COMPLETE NEST

Source of variation	df	Chi.Square	Prob.
Intercept	3	110.94	0.0000***
Exp	3	18.10	0.0004***
Site[Exp]	6	54.10	0.0000***
Antenna	3	3.66	0.3001
Year (Antenna)	15	78.05	0.0000***
Exp*Antenna	3	1.92	0.5895
Likelihood Ratio	51	91.30	0.0005***

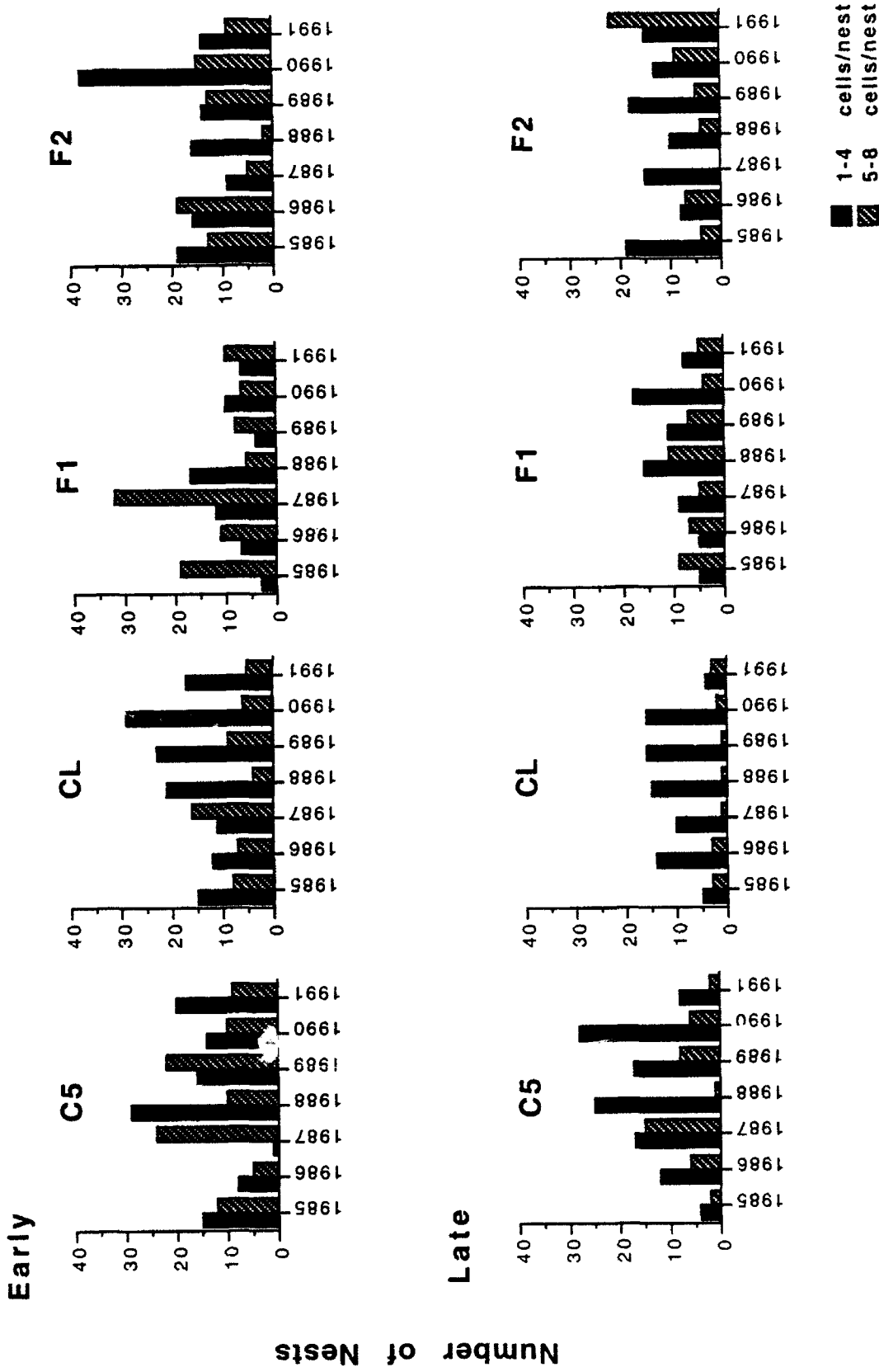


FIGURE 20 Number of complete nests of *M. relativa* with few (1-4) or many (5-12) cells, separated by season.

TABLE 15: Categorical modeling of number of cells per complete nest of Megachile relativa by season, 1985-1991.

NUMBER OF CELLS PER COMPLETE NEST			
Source of variation	df	Chi.Square	Prob.
Intercept	1	113.40	0.0000***
Exp	1	23.83	0.0000***
Site[Exp]	2	32.18	0.0000***
Antenna	1	3.63	0.0568
Year (Antenna)	5	54.20	0.0000***
Early vs. Late Season	1	26.83	0.0000***
Exp*Antenna	5	1.48	0.2237
Exp*Season	1	3.60	0.0578
Antenna*Season	1	1.63	0.2023
Exp*Antenna*Season	1	0.01	0.9233
Likelihood Ratio	41	101.67	0.0000***

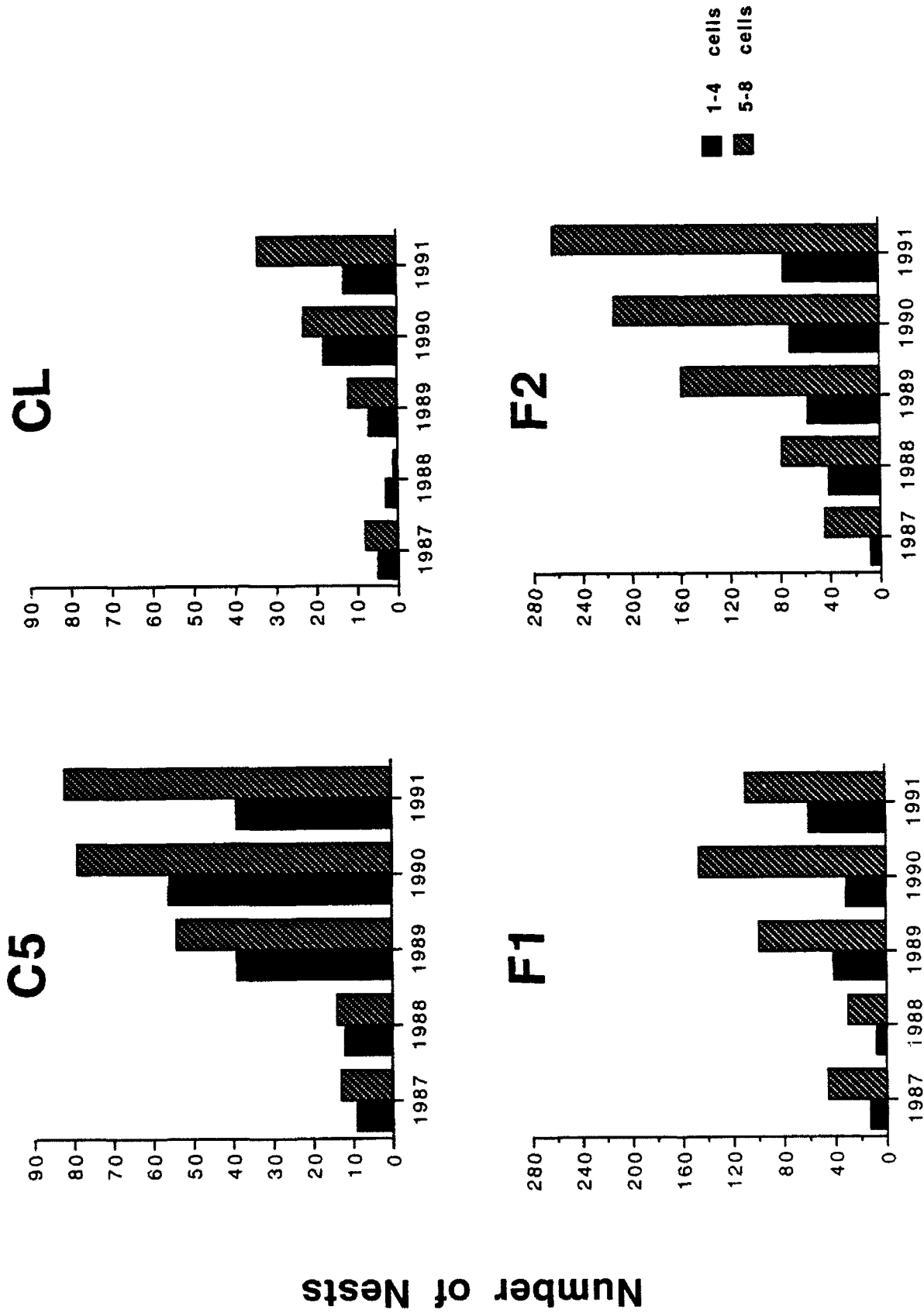


FIGURE 21. Number of complete nests of *M. inermis* with few (1-4) or many (5-8) cells, diameters >9.5 mm, bore depths >135 mm.

TABLE 16: Categorical modeling of number of cells per complete nest of Megachile inermis, 1987-1991: (diameters > 9.5mm, bore depths > 135mm).

NUMBER OF CELLS PER COMPLETE NEST

Source of variation	df	Chi.Square	Prob.
Intercept	1	31.47	0.0000***
Exp	1	18.81	0.0000***
Site[Exp]	2	0.62	0.7343
Antenna	1	6.11	0.0134**
Year (Antenna)	3	7.69	0.0529
Exp*Antenna	1	1.11	0.2927
Likelihood Ratio	11	25.83	0.0069*

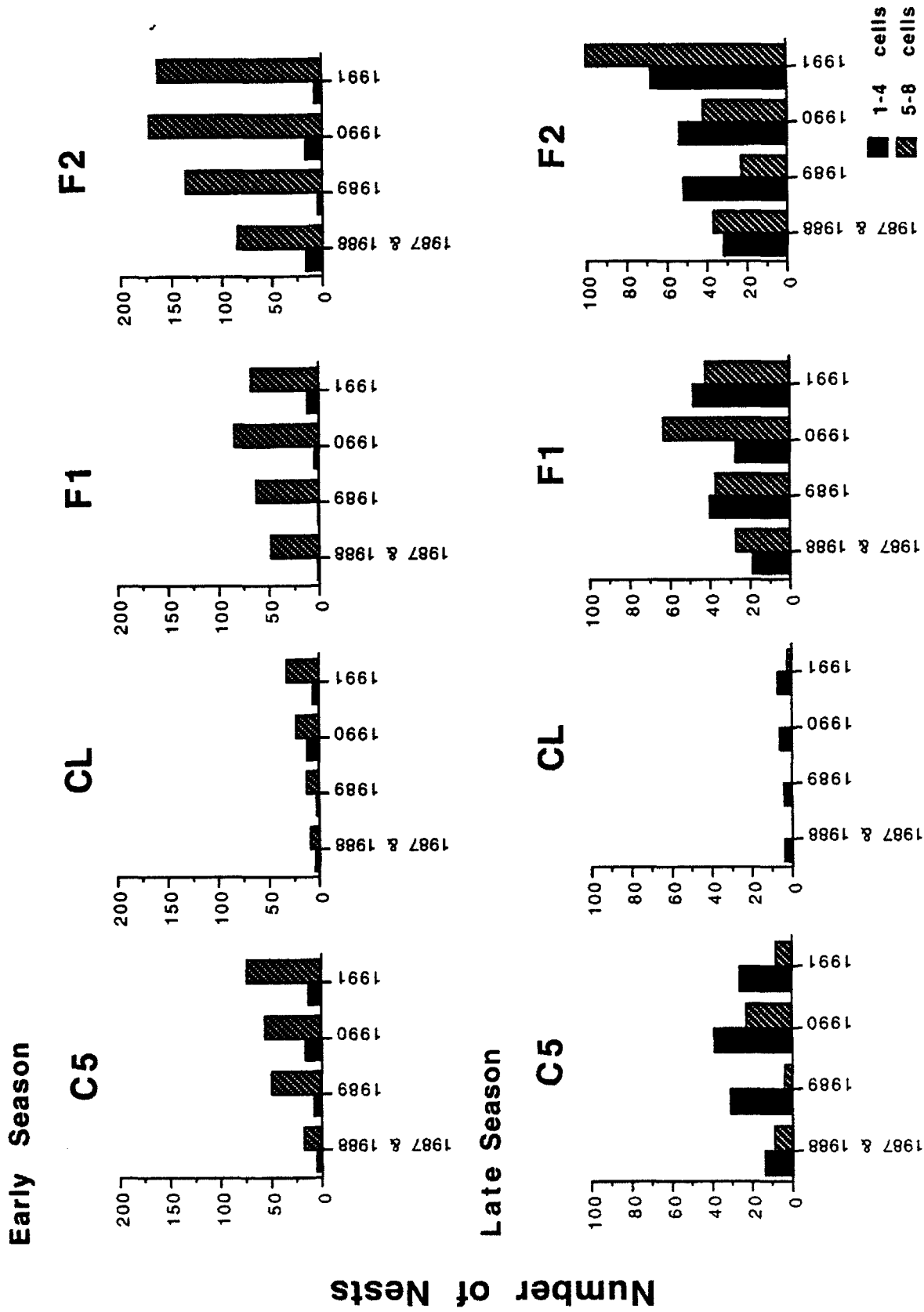


FIGURE 22. Number of complete nests of *M. inermis* with few (1-4) or many (5-8) cells, separated by season; diameters >9.5 mm, bore depths >135 mm.

TABLE 17: Categorical modeling of number of cells per complete nest of Megachile inermis, by season, 1987 + 1988 vs. 1989, 1990, 1991; diameters > 9.5mm, bore depths > 135mm.

NUMBER OF CELLS PER COMPLETE NEST

Source of variation	df	Chi.Square	Prob.
Intercept	1	62.34	0.0000***
Exp	1	73.29	0.0000***
Site[Exp]	2	8.15	0.0170*
Year (87+88 vs 89 vs 90 vs 91)	3	1.14	0.7674
Early vs. Late Season	1	287.89	0.0000***
Exp*Year	3	3.36	0.3387
Exp*Season	1	0.07	0.7913
Year*Season	3	24.22	0.0000***
Exp.*Year*Season	3	3.29	0.3492
Likelihood Ratio	14	41.53	0.0001***

Nest Plug Length *Megachile inermis*

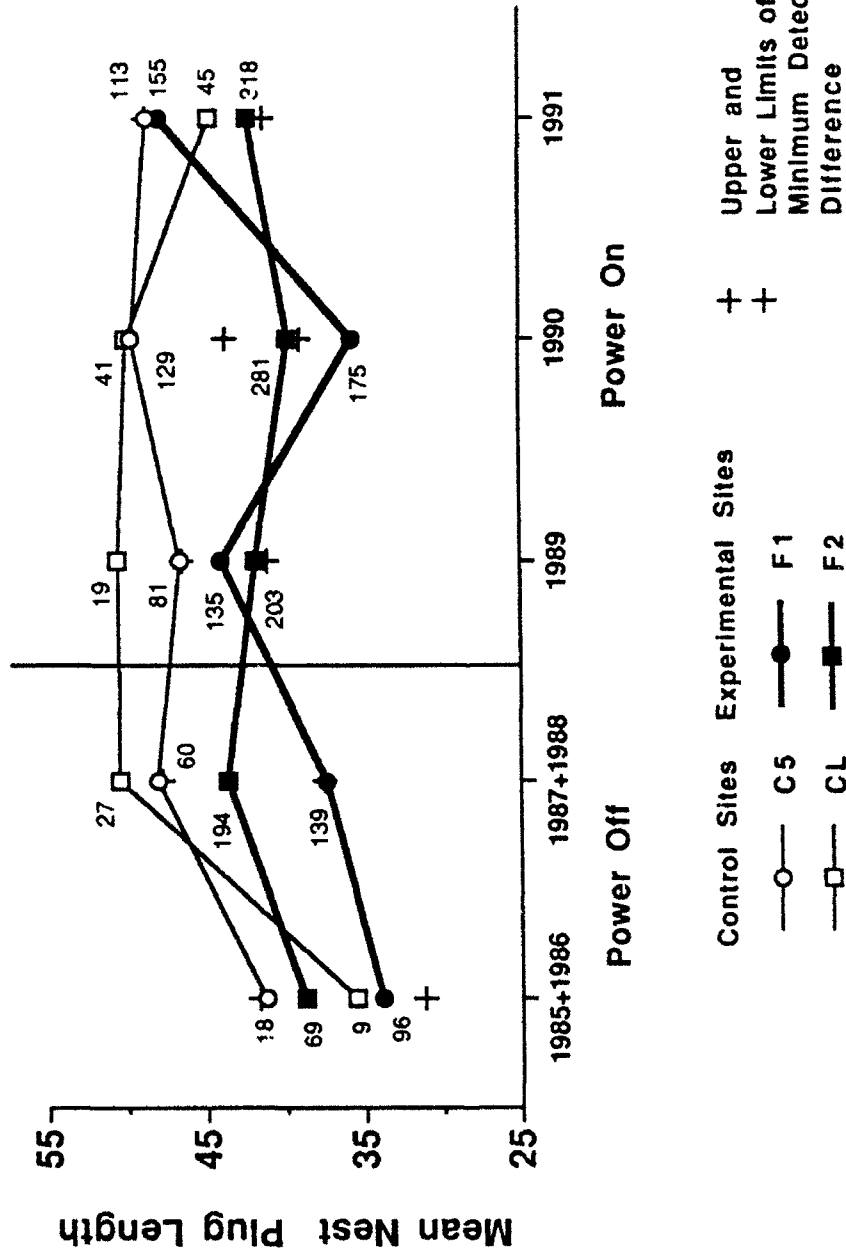


FIGURE 23. Mean nest plug lengths for *M. inermis*; diameters >9.5. Numbers indicate sample sizes; + indicate upper and lower limits to the minimum detectable difference between experimental and control sites for each year.

TABLE 18: GLM of *M. inermis* nest plug lengths from 1985-1991; diameters > 9.5 mm.

PLUG LENGTHS

Source of variation	Numerator df	MS	F	P>F
Exp	1	3130.69	19.28	0.0001***
Site [Exp]	2	52.78	0.20	0.8165
Antenna	1	2865.31	1.20	0.3456
Year [Antenna] 85+86, 87+88, 89, 90, 91	3	5089.06	13.98	0.0002***
Measurer [Year * Antenna]	12	370.69	1.42	0.1471
Exp * Antenna	1	147.98	0.57	0.4509
Diameter	1	1968.95	7.57	0.0060*
Cells per nest	1	143418.87	551.11	0.0001***
Early vs. Late Season	1	4187.46	16.09	0.0001***
Model	23	8500.41	32.66	0.0001***
Error	2283	260.24		
$\bar{X} = 42.66$ mm	CV=37.82	$r^2=0.25$		

TABLE 18 (continued)

Parameter	Estimate:	T for H_0 : Parameter = 0	$PR > T $	\bar{X}	SD
Exp:					
Control Sites	3.08	2.96	0.0031*	48.02	19.97
Experimental Sites	0.0	—	—	41.01	17.71
Year [Antenna]:					
Off: 85+86	-13.28	-4.96	0.0001***	36.47	17.61
87+88	0.0	—	—	42.69	17.52
On: 89	2.07	1.19	0.2342	43.83	19.36
90	-3.08	-1.95	0.0516	41.34	18.11
91	0.0	—	—	45.02	18.70
Diameter	2.17	2.75	0.0060*		
Cells per nest	-6.43	-23.48	0.0001***		
Early Season vs. Late Season	3.15 0.0	4.01 —	0.0001*** —		

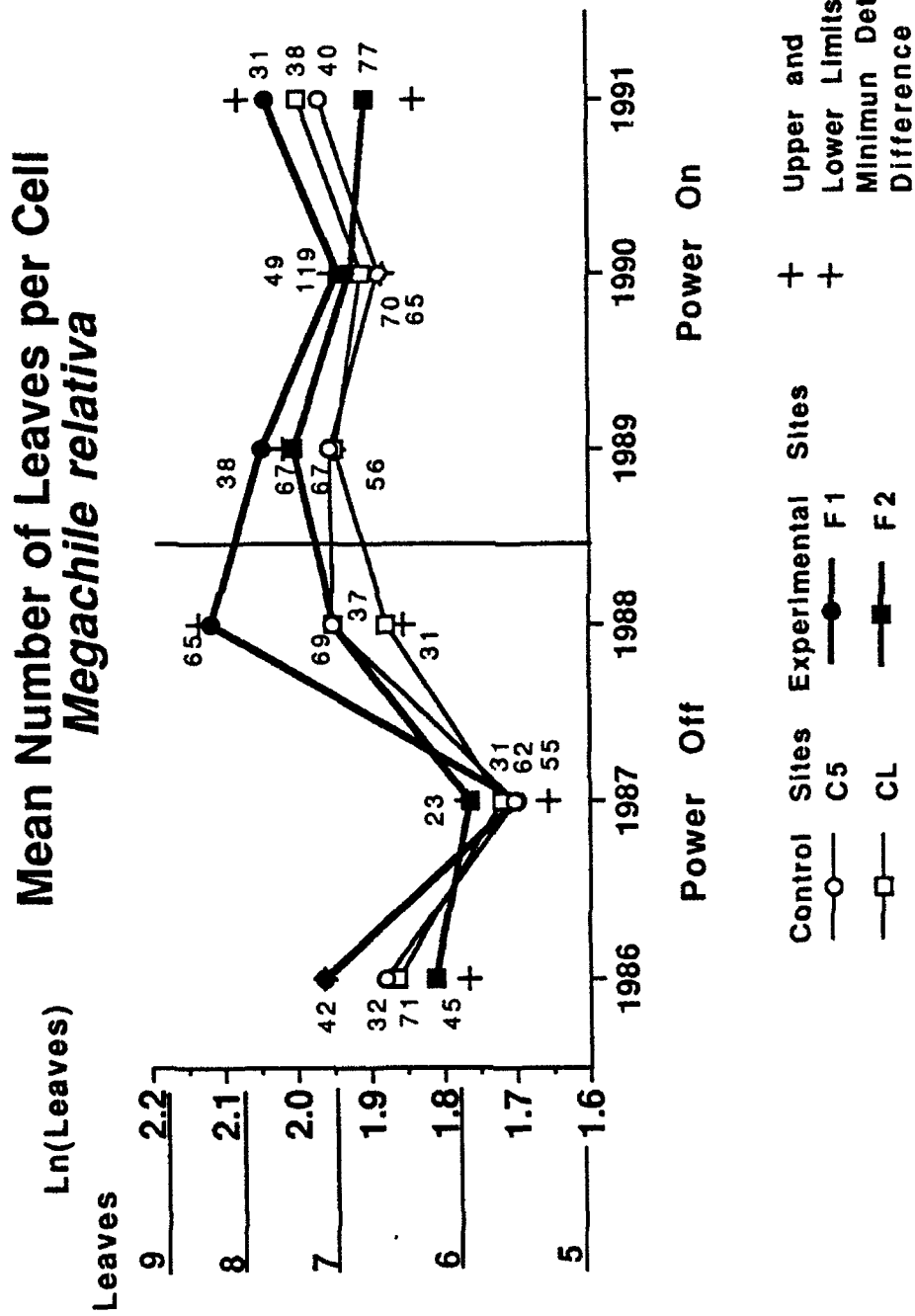


FIGURE 24. Mean leaves per cell for *M. relativa* nests. Numbers indicate sample sizes; + indicate the minimum detectable difference between experimental and control sites for each year.

TABLE 19: GLM of mean of ln transformed leaves per cell for all cells from 1986-1991 *M. relativa* nests.

LEAVES PER CELL

Source of variation	Numerator df	MS	F	P>F
Exp	1	0.65	1.67	0.3262
Site [Exp]	2	0.38	10.96	0.0001***
Antenna	1	1.17	0.87	0.4042
Year [Antenna]	4	1.46	41.77	0.0001***
Exp * Antenna	1	0.00	0.07	0.7992
Diameter	1	7.76	222.06	0.0001***
Complete vs. incomplete	1	0.07	1.97	0.1612
Cells per nest	1	0.07	2.11	0.1462
Early vs. Late Season	1	1.32	37.75	0.0001***
Model	13	1.57	45.00	0.0001***
Error	1266	0.03		
$\bar{X} = 1.91$ (6.8 leaves)	CV=9.8	$r^2=0.32$		

TABLE 19 (continued)

Parameter	Estimates:	T for H ₀ : Parameter = 0	PR> T	\bar{X}	SD
Sites [Exp]					
C5	0.0	--	--	1.89	0.21
CL	0.001	0.04	0.9656	1.90	0.23
F1	0.073	4.68	0.0001***	1.96	0.24
F2	0.0	--	--	1.91	0.22
Year (Antenna)					
Off: 1986	-0.079	-4.04	0.0001***	1.87	0.25
1987	0.242	-12.02	0.0001***	1.71	0.16
1988	0.0	--	--	1.99	0.22
On: 1989	0.035	1.88	0.0604	1.98	0.21
1990	-0.030	-1.73	0.084	1.92	0.19
1991	0.0	--	--	1.96	0.21
Diameter	0.168	14.90	0.0001***		
Early Season vs. Late Season	-0.067 0.0	-6.14 --	0.0001*** --		

¹ Ln (Leaves per Cell)

TABLE 20: GLM of mean of ln transformed leaves per cell in *M. relativa* nests; expected sex included in the model.

LEAVES PER CELL

Source of variation	Numerator df	MS	F	P>F
Exp	1	0.21	0.61	0.5125
Site [Exp]	2	0.46	13.59	0.0001***
Antenna	1	0.60	0.60	0.4806
Year [Antenna]	4	1.41	41.57	0.0001***
Exp * Antenna	1	0.06	1.62	0.2028
Sex	1	0.74	21.88	0.0001***
Exp * Sex	1	0.00	0.00	0.9563
Antenna * Sex	1	0.00	0.09	0.7637
Exp * Antenna * Sex	1	0.02	0.51	0.4750
Diameter	1	6.53	191.93	0.0001***
Complete vs. incomplete	1	0.01	0.17	0.6827
Cells per nest	1	0.02	0.55	0.4596
Early vs. Late Season	1	0.68	19.88	0.0001***
Model	17	1.08	31.75	0.0001***
Error	1022	0.03		
$\bar{X} = 1.90$ (6.7 leaves) $CV = 9.7$ $r^2 = 0.35$				

TABLE 20 (continued)

Parameter	Estimate	T for H ₀ :		\bar{X}^1	SD
		Parameter = 0	PR > T		
Sites [Exp]					
C5	0.0	--	--	1.88	0.21
CL	0.003	0.16	0.8729	1.90	0.22
F1	0.087	5.20	0.0001***	1.95	0.24
F2	0.0	--	--	1.90	0.23
Year [Antenna]					
Off 1986	-0.114	-5.08	0.0001***	1.85	0.26
1987	-0.269	-11.74	0.0001***	1.71	0.15
1988	0.0	--	--	2.02	0.22
On 1989	-0.054	2.71	0.0068*	1.99	0.21
1990	-0.034	-1.78	0.0751	1.90	0.19
1991	0.0	--	--	1.95	0.19
Sex F	-0.079	-3.11	0.0019*	1.87	0.22
M	0.0	--	--	1.92	0.23
Diameter	0.166	13.85	0.0001***		
Early vs.	-0.055	-4.46	0.0001***		
Late Season	0.0	--	--		

¹ Ln (Leaves per cell)

Mean Number of Leaves per Cell *Megachile inermis*

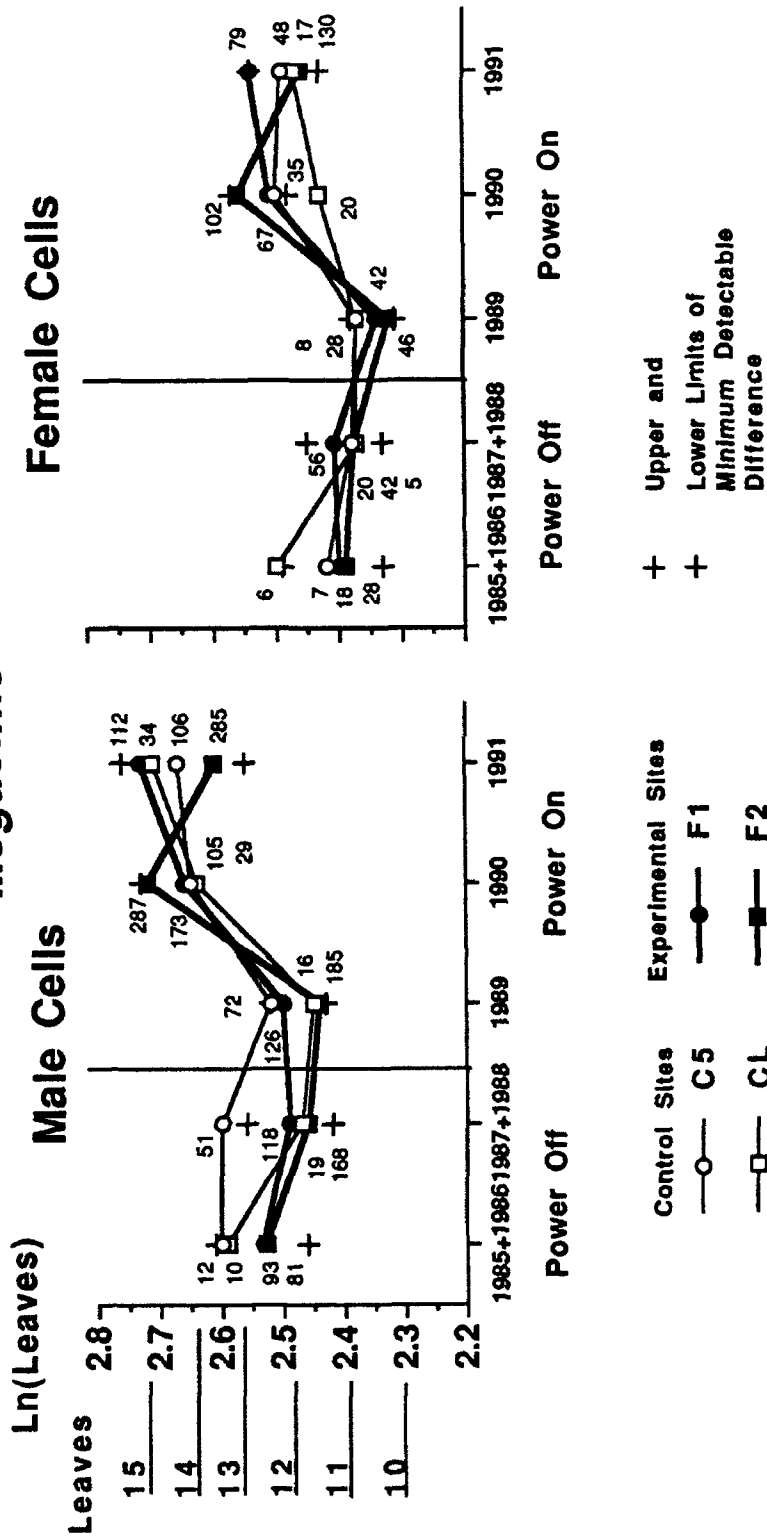


FIGURE 25. Mean leaves per cell for *M. inermis* nests by sex; diameters >9.5mm. Numbers indicate sample sizes; + indicate the minimum detectable difference between experimental and control sites for each year.

TABLE 21: GLM of mean of ln transformed leaves per cell in *M. inermis* nests; diameters >9.5mm; expected sex included in the model.

LEAVES PER CELL

Source of variation	Numerator df	MS	F	P>F
Exp	1	0.18	2.62	0.1821
Site [Exp]	2	0.13	4.22	0.0148*
Antenna	1	0.02	0.03	0.8645
Year [Antenna] 85+86, 87+88, 89, 90, 91	3	1.39	44.58	0.0001***
Exp * Antenna	1	0.23	7.46	0.0063*
Sex	1	5.70	182.63	0.0001***
Exp * Sex	1	0.11	3.41	0.0650
Antenna * Sex	1	0.04	1.37	0.2415
Exp * Antenna * Sex	1	0.05	1.75	0.1862
Diameter	1	14.49	464.36	0.0001***
Complete vs. incomplete	1	0.27	8.64	0.0033*
Cells per nest	1	0.34	10.94	0.0010**
Early vs. Late Season	1	4.21	134.97	0.0001***
Model	16	3.25	104.30	0.0001***
Error	2872	89.61		
$\bar{X} = 2.55$ (12.8 leaves) $CV = 6.9$ $r^2 = 0.37$ For $\alpha=0.05$ Power of Exp * Antenna=0.78				

TABLE 21 (continued)

Parameter	Estimate	T for H ₀ : Parameter = 0	PR > T	\bar{X}^1	SD
Exp * Antenna					
Control Off	0.087	3.79	0.0002	2.52	0.23
Control On	0.0	--	--	2.57	0.21
Experimental Off	0.0	--	--	2.47	0.22
Experimental On	0.0	--	--	2.58	0.22
Sites [Exp]					
C5	0.0	--	--	2.57	0.21
CL	-0.015	-0.93	0.3548	2.54	0.22
F1	0.021	2.76	0.0058	2.55	0.23
F2	0.0	--	--	2.55	0.22
Year [Antenna]					
Off 1985 + 1986	0.029	2.02	0.0435*	2.51	0.25
1987 + 1988	0.0	--	--	2.47	0.20
On 1989	-0.017	-1.42	0.1549	2.44	0.16
1990	0.083	9.16	0.0001***	2.64	0.21
1991	0.0	--	--	2.60	0.21
Sex F	-0.152	-15.46	0.0001***	2.45	0.19
M	0.0	--	--	2.59	0.22
Diameter	0.163	21.55	0.0001		
Complete vs. Incomplete	-0.04 0.0	-2.94 --	0.0033* --		
Cells per nest	-0.010	-3.31	0.0010**		
Early vs. Late Season	-0.090 0.0	-11.62 --	0.0001*** --		

¹ Ln (Leaves per cell)

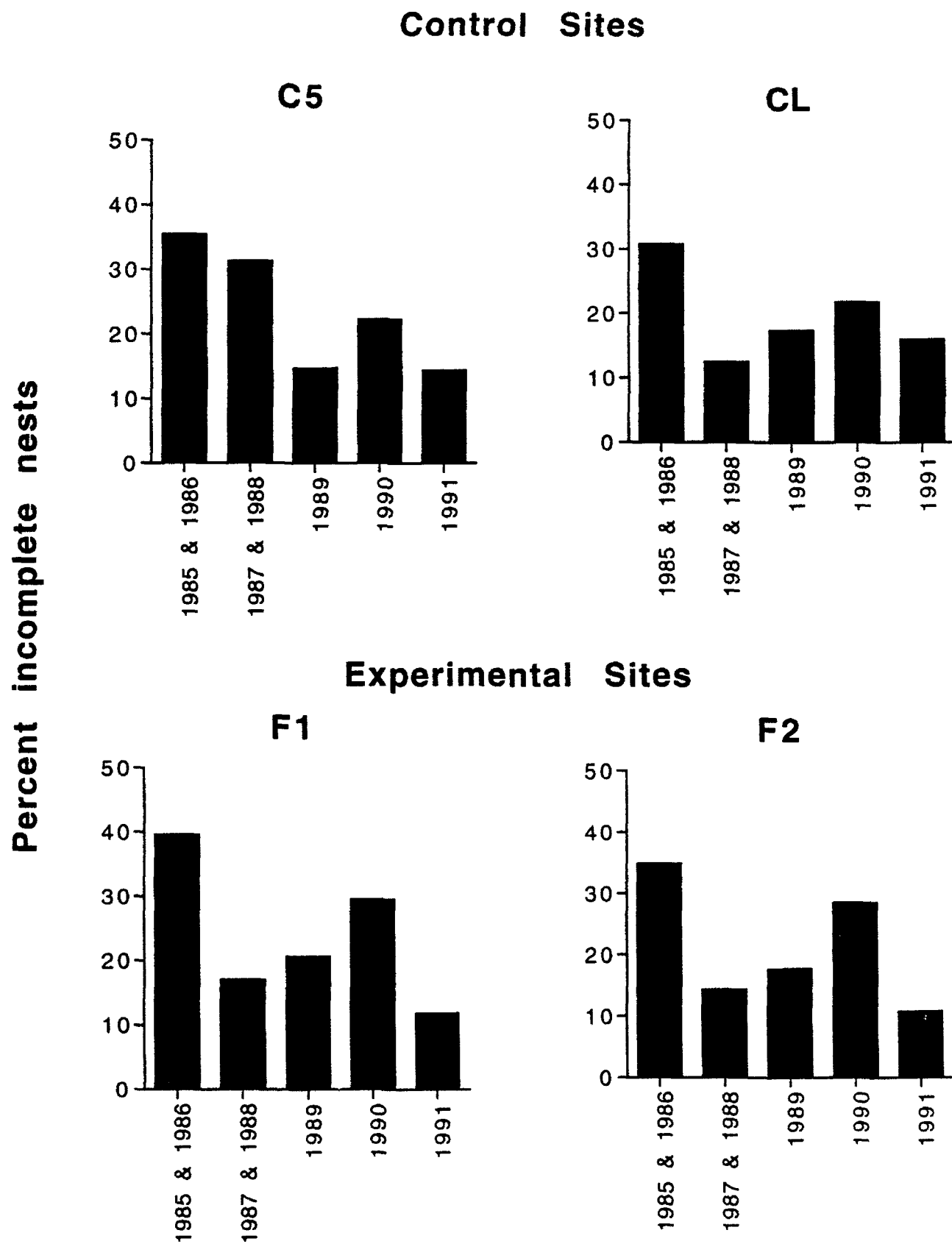


FIGURE 26. Percent incomplete and usurped nests for *M. inermis* by site and year.

TABLE 22: LOG-LIKELIHOOD RATIO CONTINGENCY TABLES FOR *M. RELATIVA* NEST ENTRANCE ORIENTATION BY HUTCH SET AND YEAR.

H_0 : Nest orientations at each hutch set are homogeneous between years (i.e., have the same directional preference).

H_1 : Nest orientations at each hutch set are heterogeneous between years and hutch sets at a site, so data cannot be pooled.

	EW	NS	R		EW	NS	R		
C5-S					CL-E				
1983	—	—	—	G=8.688 df = 6 n.s.	14	9	23	G=4.904 df=7 n.s.	
1985	6	5	11		15	9	24		
1986	4	6	10		8	3	11		
1987	6	16	22		12	6	18		
1988	4	19	23		13	2	15		
1989	9	12	21		12	7	19		
1990	12	18	30		13	10	23		
1991	3	13	16	4	2	6			
C	44	89	133		91	48	139		
C5-N					CL-N				
1983	—	—	—	G=6.623 df=5 n.s.	—	—	—	G=7.945 df=6 n.s.	
1985	4	2	6		12	7	19		
1986	5	6	11		10	7	17		
1987	4	3	7		10	3	13		
1988	12	3	15		17	4	21		
1989	9	13	22		11	1	12		
1990	10	8	18		10	1	11		
1991	1	4	5*	7	2	9			
C	44	35	79		77	25	102		
C5-W					CL-W				
1983	—	—	—	G=10.798 df=6 n.s.	—	—	—	G=3.033 df=6 n.s.	
1985	8	14	22		7	4	11		
1986	11	7	18		6	8	14		
1987	18	18	36		5	6	11		
1988	14	24	38		3	6	9		
1989	14	10	24		9	11	20		
1990	8	6	14		12	14	26		
1991	16	6	22	9	6	15			
C	89	85	174		51	55	106		
C5 - BY HUTCH SETS					CL - BY HUTCH SETS				
C5-S	44	89	133	G=13.994 ¹ df=2 P<.001	CL-E	91	48	139	G=17.325 ¹ df=2 P<.001
C5-N	44	35	79		CL-N	77	25	102	
C5-W	89	85	174		CL-W	51	55	106	
C	178	213	391			219	128	347	

1. Within hutch sets, data are homogeneous between years. However, hutch sets (data pooled across years) are heterogeneous. Thus, hutch set data cannot be pooled by year.

* C5:N hutches were moved late summer 1990; 1990 nests are included in analyses because most nests were constructed before the hutches were moved. 1991 nests were not included in analyses.

TABLE 22 (continued)

H_0 : Nest orientations at each hutch set are homogeneous between years (i.e., have the same directional preference).

H_1 : Nest orientations at each hutch set are heterogeneous between years and hutch sets at a site, so data cannot be pooled.

	EW	NS	R		EW	NS	R	
F1-E				F2-E				
1983	42	25	67	—	—	—		
1985	15	6	21	9	5	14		
1986	12	4	16	10	16	26	$G=10.235^1$	
1987	18	21	39	6	9	15	$df=6$	
1988	7	9	16	4	16	20	n.s.	
1989	5	6	11	10	12	22		
1990	5	7	12	6	20	26		
1991	5	4	9	8	17	25		
	109	82	191	53	95	148		
F1-N				F2-N				
1983	—	—	—	—	—	—		
1985	15	5	20	20	17	37**		
1986	5	8	13	10	23	33**		
1987	6	16	22	7	10	17	$G=8.638$	
1988	4	22	26	5	8	13	$df=4^*$	
1989	2	16	18	19	8	27	n.s.	
1990	6	9	15	28	12	40		
1991	4	9	13	10	10	20		
	42	85	127	69	48	117		
F1-W				F2-W				
1983	21	21	42	—	—	—		
1985	2	12	14	8	10	18		
1986	4	2	6	5	1	6	$G=11.638^1$	
1987	2	2	4	2	4	6	$df=6$	
1988	10	5	15	3	3	6	n.s.	
1989	2	2	4	3	4	7		
1990	1	12	13	14	6	20		
1991	5	5	10	5	14	19		
	47	61	108	40	42	82		
F2 - BY HUTCH SETS								
F2-E	53	95	148					
F2-N	69	48	117				$G=14.397^1$	
F2-W	40	42	82				$df=2$	
	162	185	347				$P<.001$	

1. Within hutch sets, data are homogeneous between years. However, hutch sets (data pooled across years) are heterogeneous. Thus, hutch set data cannot be pooled by year.

2. Within hutch sets data are heterogeneous; cannot be pooled.

** Hutchers were moved in spring, 1987, so 1985 & 1986 were not included in analyses.

Mean Duration of LO Collecting Trips *Megachile inermis*

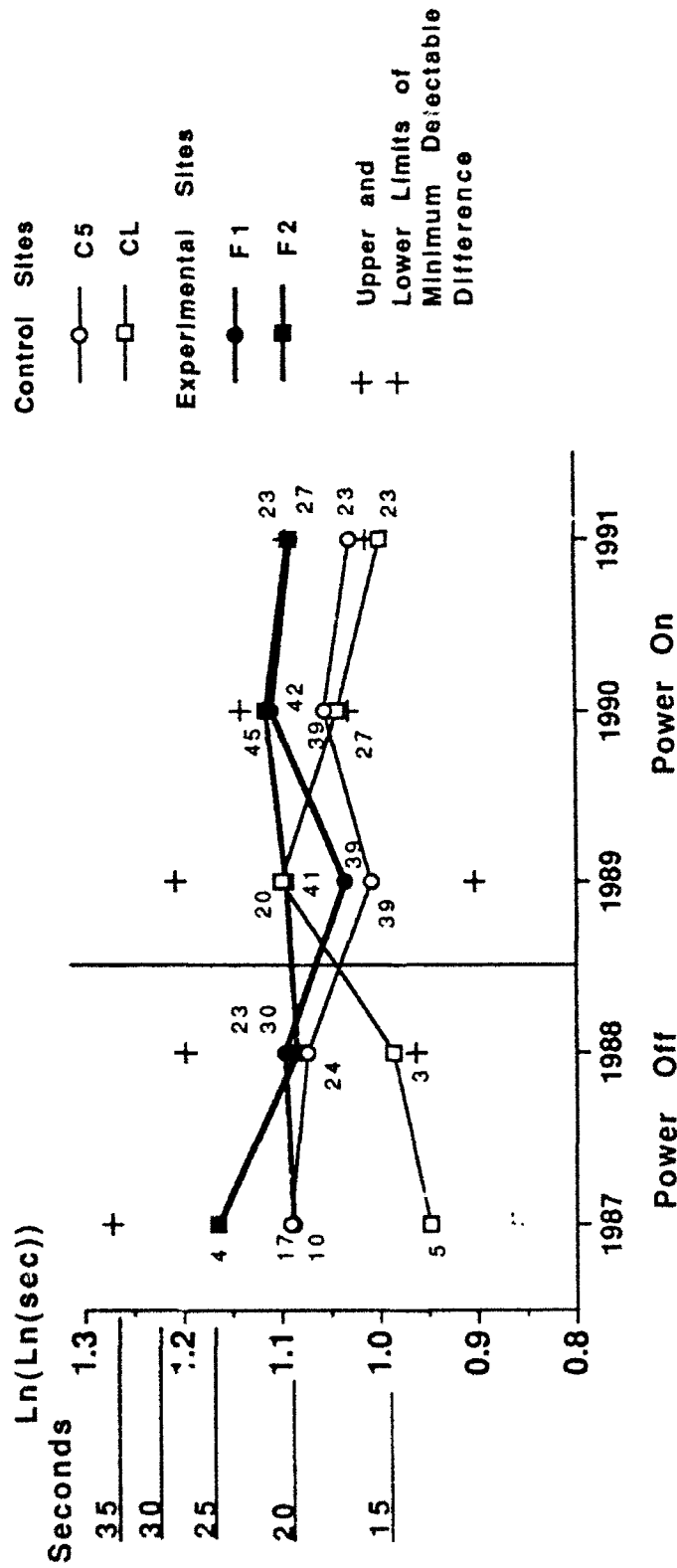


FIGURE 27. Mean duration of LO collecting trips for the first three leaf collecting trips in a cell cap. Numbers indicate number of bees timed; + indicate upper and lower limits to the minimum detectable difference between experimental and control sites for each year.

TABLE 23: GLM of mean of ln ln transformed LO trip durations; trips 1-3 for each timed M. inermis; "OFF" vs. "ON" antenna operation.

MEAN LO TRIP DURATIONS

Source of variation	Numerator df	MS	F	P>F
Exp	1	0.40	6.19	0.0355*
Site [Exp]	2	0.06	0.72	0.4854
Antenna	1	0.00	0.00	0.9884
Year [Antenna]	3	0.05	0.65	0.5806
Observer [Year * Antenna]	17	0.05	0.65	0.8556
Exp * Antenna	1	0.01	0.09	0.7609
Time [Year * Antenna]	5	0.05	0.62	0.6856
Time ² [Year * Antenna]	5	0.05	0.65	0.6647
Date [Year * Antenna]	5	0.26	3.45	0.0045*
Model	40	0.13	1.71	0.0057*
Error	463	0.08		
$\bar{X} = 1.07$ (18.4 sec.)	CV = 25.8	$r^2 = 0.13$		

Parameter	Estimate	T for H ₀ : Parameter = 0	PR> T
Exp: Control Sites	-0.053	-1.21	0.2274
Experimental Sites	0.0	--	--
Date [Year * Antenna]			
1987	0.002	0.83	0.4081
1988	0.002	1.17	0.2440
1989	0.002	1.57	0.1165
1990	0.003	3.49	0.0005**
1991	0.001	0.81	0.4167

TABLE 24: *M. relativa* sex ratio by site and year.

Site	1985			1986		
	Males	Females	Ratio	Males	Females	Ratio
C5	98	9	10.9	69	23	3.0
CL	129	49	2.6	75	9	8.3
F1	262	42	6.2	94	18	5.2
F2	129	30	4.3	204	32	6.4
Total	618	130	4.8	442	82	5.4

Site	1987			1988		
	Males	Females	Ratio	Males	Females	Ratio
C5	207	67	3.1	70	25	2.8
CL	55	24	2.3	23	7	3.3
F1	186	60	3.1	111	12	9.3
F2	38	7	5.4	32	9	3.6
Total	486	158	3.1	236	53	4.5

Site	1989			1990		
	Males	Females	Ratio	Males	Females	Ratio
C5	148	70	2.1	125	26	4.8
CL	54	35	1.5	78	16	4.9
F1	95	18	5.3	92	26	3.5
F2	101	21	4.8	221	44	5.0
Total	398	144	2.8	516	112	4.6

Site	1991		
	Males	Females	Ratio
C5	61	22	2.8
CL	37	12	3.1
F1	81	34	2.4
F2	186	28	6.6
Total	365	96	3.8

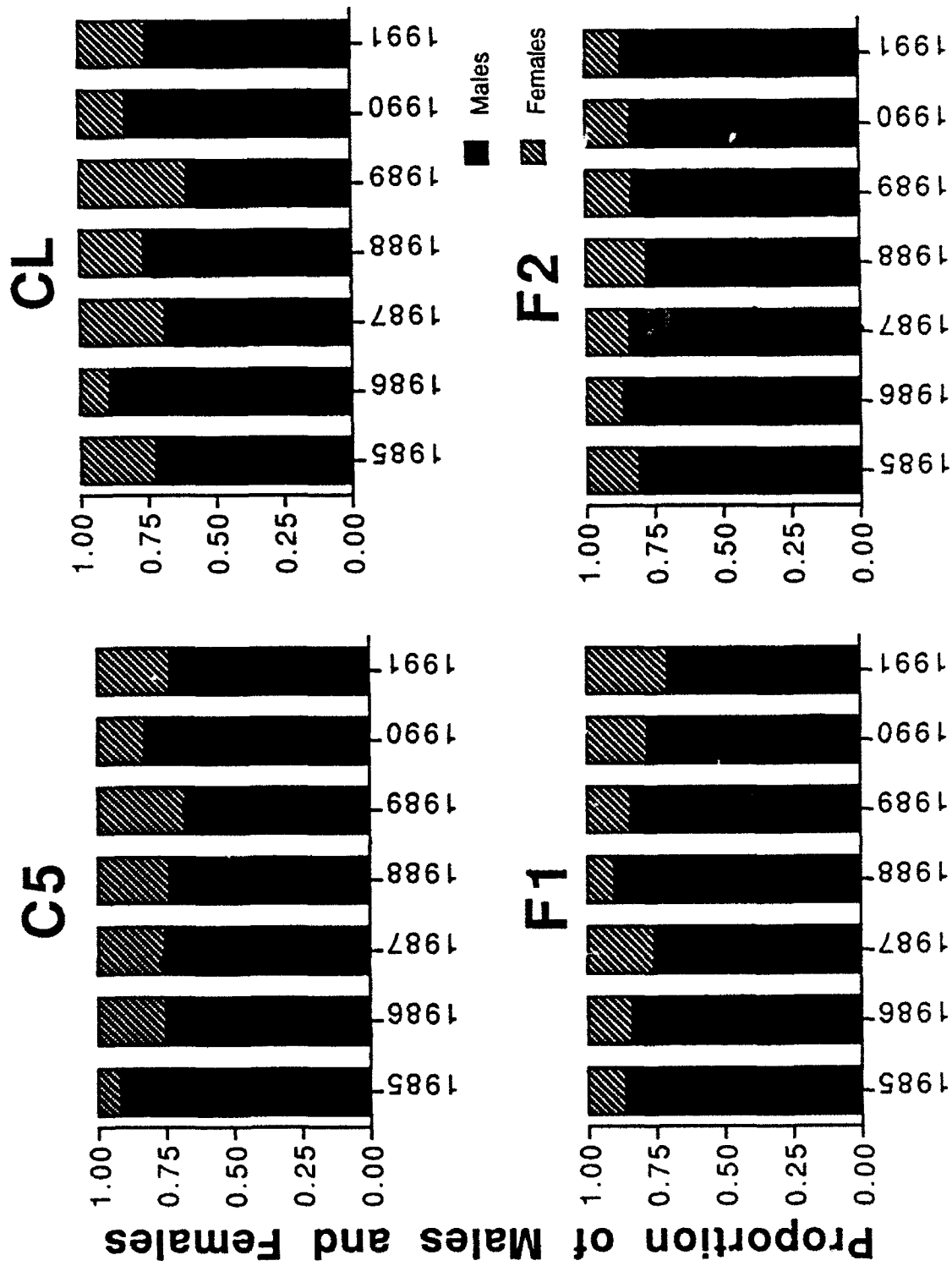


FIGURE 28. Relative proportions of male and female *M. relativa* by site and year.

TABLE 25: Categorical modeling of male and female emergences for Megachile relativa, 1985-1991.

SEX RATIO

Source of variation	df	Chi.Square	Prob.
Intercept	1	250.75	0.0000***
Exp	1	23.73	0.0000***
Site [Exp]	2	3.88	0.1440
Antenna	1	0.00	0.9757
Year [Antenna]	5	21.90	0.0005***
Exp*Antenna	1	0.25	0.6191
Likelihood Ratio	17	60.37	0.0000***

TABLE 26: Late Summer Emergences (% bivoltinism) of M. relativa and Coelioxys spp.

Year	<u>M. relativa</u>			
	cells emerging late summer	total cells emerging ¹ (%)	nests emerging late summer	total nests emerging ¹ (%)
1987	33/629	(5.2%)	7/186	(3.8%)
1988	13/285	(4.6%)	7/144	(4.9%)
1989	112/515	(21.7%)	24/166	(14.5%)
1990	41/621	(6.6%)	13/232	(5.6%)
1991	26/452	(5.8%)	7/165	(4.2%)

Year	<u>Coelioxys</u> spp.			
	cells emerging late summer	total cells emerging ² (%)	nests emerging late summer	total nests emerging ² (%)
1987	11/99	(11.1%)	10/77	(13.0%)
1988	10/87	(11.5%)	8/62	(12.9%)
1989	18/71	(25.4%)	11/50	(22.0%)
1990	7/116	(6.0%)	6/92	(6.5%)
1991	12/107	(11.2%)	12/75	(16.0%)

¹Total cells or nests with adult M. relativa.

²Total cells or nests with adult Coelioxys in M. relativa nests.

TABLE 27: Late Summer Emergences (% bivoltinism) of M. inermis and Coelioxys spp.

<u>M. inermis</u>						
Year	cells emerging late summer	total cells emerging ¹	(%)	nests emerging late summer	total nests emerging ¹	(%)
1987	2/1011		(0.2%)	1/262		(0.4%)
1988	0/562		(0.0%)	0/168		(0.0%)
1989	4/1190		(0.3%)	1/400		(0.3%)
1990	5/1969		(0.3%)	1/628		(0.2%)
1991	4/2160		(0.2%)	1/606		(0.2%)

<u>Coelioxys</u> spp.						
Year	cells emerging late summer	total cells emerging ²	(%)	nests emerging late summer	total nests emerging ²	(%)
1987	0/62		(0.0%)	0/48		(0.0%)
1988	0/18		(0.0%)	0/16		(0.0%)
1989	0/86		(0.0%)	0/67		(0.0%)
1990	0/85		(0.0%)	0/70		(0.0%)
1991	0/51		(0.0%)	0/48		(0.0%)

¹Total cells or nests with adult M. inermis.²Total cells or nests with adult Coelioxys in M. inermis nests.

TABLE 28: Proportion of M. relativa mortality from various sources by site.

Stage or source of mortality	SITE			
	C5	CL	F1	F2
1983				
Pre-overwintering (egg & larvae)			0.149	
Overwintering (Prepupae)			0.117	
Total parasitism (<u>Coelioxys</u> only)			0.102 (0.063)	
Post-overwintering Survival*			0.632	
1985				
Pre-overwintering (egg & larvae)	0.185	0.131	0.059	0.053
Overwintering (Prepupae)	0.045	0.069	0.014	0.041
Total parasitism (<u>Coelioxys</u> only)	0.089 (0.076)	0.073 (0.053)	0.100 (0.089)	0.254 (0.234)
Post-overwintering Survival*	0.681	0.727	0.827	0.652
1986				
Pre-overwintering (egg & larvae)	0.104	0.138	0.109	0.041
Overwintering (Prepupae)	0.130	0.015	0.085	0.063
Total parasitism (<u>Coelioxys</u> only)	0.169 (0.130)	0.177 (0.138)	0.127 (0.127)	0.149 (0.114)
Post-overwintering Survival*	0.597	0.669	0.679	0.749
1987				
Pre-overwintering (egg & larvae)	0.235	0.354	0.186	0.344
Overwintering (Prepupae)	0.041	0.030	0.055	0.070
Total parasitism (<u>Coelioxys</u> only)	0.058 (0.041)	0.128 (0.122)	0.118 (0.110)	0.234 (0.195)
Post-overwintering Survival*	0.665	0.488	0.640	0.352

* Includes cells with dead pupae, dead adults, and successfully emerging adult M. relativa.

TABLE 28 (continued)

Stage or source of mortality	C5	CL	SITE F1	F2
1988				
Pre-overwintering (egg & larvae)	0.313	0.407	0.363	0.464
Overwintering (Prepupae)	0.134	0.122	0.106	0.064
Total parasitism (<u>Coelioxys</u> only)	0.167 (0.138)	0.228 (0.195)	0.099 (0.070)	0.144 (0.128)
Post-overwintering Survival*	0.386	0.244	0.433	0.328
1989				
Pre-overwintering (egg & larvae)	0.106	0.127	0.080	0.176
Overwintering (Prepupae)	0.165	0.127	0.105	0.188
Total parasitism (<u>Coelioxys</u> only)	0.083 (0.071)	0.206 (0.139)	0.117 (0.111)	0.130 (0.092)
Post-overwintering Survival*	0.646	0.539	0.698	0.506
1990				
Pre-overwintering (egg & larvae)	0.095	0.201	0.179	0.116
Overwintering (Prepupae)	0.069	0.082	0.067	0.067
Total parasitism (<u>Coelioxys</u> only)	0.182 (0.147)	0.207 (0.179)	0.101 (0.101)	0.136 (0.105)
Post-overwintering Survival*	0.654	0.511	0.654	0.681
1991				
Pre-overwintering (egg & larvae)	0.223	0.291	0.136	0.090
Overwintering (Prepupae)	0.072	0.127	0.043	0.093
Total parasitism (<u>Coelioxys</u> only)	0.205 (0.193)	0.216 (0.142)	0.111 (0.105)	0.182 (0.146)
Post-overwintering Survival*	0.500	0.366	0.710	0.636

* Includes cells with dead pupae, dead adults, and successfully emerging adult M. relativa.

TABLE 29: Proportion of *M. inermis* mortality from various sources by site.

Stage or source of mortality	SITE			
	C5	CL	F1	F2
1985				
Pre-overwintering (egg & larvae)	0.151	0.098	0.184	0.114
Overwintering (Prepupae)	0.019	0.000	0.028	0.022
Total parasitism (<i>Coelioxys</i> only)	0.189 (0.170)	0.176 (0.059)	0.031 (0.011)	0.079 (0.035)
Post-overwintering Survival*	0.641	0.725	0.757	0.786
1986				
Pre-overwintering (egg & larvae)	0.119	0.000	0.061	0.004
Overwintering (Prepupae)	0.051	0.000	0.038	0.026
Total parasitism (<i>Coelioxys</i> only)	0.068 (0.034)	0.000 (0.000)	0.167 (0.038)	0.073 (0.009)
Post-overwintering Survival*	0.763	1.000	0.735	0.897
1987				
Pre-overwintering (egg & larvae)	0.272	0.062	0.131	0.124
Overwintering (Prepupae)	0.092	0.062	0.055	0.069
Total parasitism (<i>Coelioxys</i> only)	0.072 (0.048)	0.186 (0.088)	0.070 (0.032)	0.103 (0.041)
Post-overwintering Survival*	0.564	0.690	0.744	0.704
1988				
Pre-overwintering (egg & larvae)	0.174	0.300	0.175	0.260
Overwintering (Prepupae)	0.165	0.100	0.087	0.085
Total parasitism (<i>Coelioxys</i> only)	0.035 (0.009)	0.150 (0.150)	0.044 (0.000)	0.060 (0.025)
Post-overwintering Survival*	0.626	0.450	0.694	0.595

* Includes cells with dead pupae, dead adults, and successfully emerging adult *M. inermis*.

TABLE 29 (continued)

Stage or source of mortality	C5	SITE		F2	
		CL	F1	<u>overwintered</u>	
				C5	F2
1989					
Pre-overwintering (egg & larvae)	0.183	0.156	0.130		0.171
Overwintering (Prepupae)	0.214	0.167	0.213		0.240
Total parasitism (<u>Coelioxys</u> only)	0.060 (0.036)	0.156 (0.115)	0.099 (0.042)		0.062 (0.032)
Post-overwintering Survival*	0.542	0.521	0.558		0.527
1990					
Pre-overwintering (egg & larvae)	0.206	0.249	0.137	0.074	0.069
Overwintering (Prepupae)	0.159	0.180	0.186	0.164	0.214
Total parasitism (<u>Coelioxys</u> only)	0.091 (0.042)	0.152 (0.106)	0.078 (0.013)	0.057 (0.021)	0.083 (0.028)
Post-overwintering Survival*	0.543	0.419	0.599	0.706	0.634
1991					
Pre-overwintering (egg & larvae)	0.151	0.258	0.200	0.105	0.144
Overwintering (Prepupae)	0.072	0.146	0.079	0.074	0.089
Total parasitism (<u>Coelioxys</u> only)	0.057 (0.007)	0.100 (0.029)	0.090 (0.017)	0.059 (0.014)	0.074 (0.023)
Post-overwintering Survival*	0.719	0.496	0.630	0.761	0.693

* Includes cells with dead pupae, dead adults, and successfully emerging adult M. inermis.

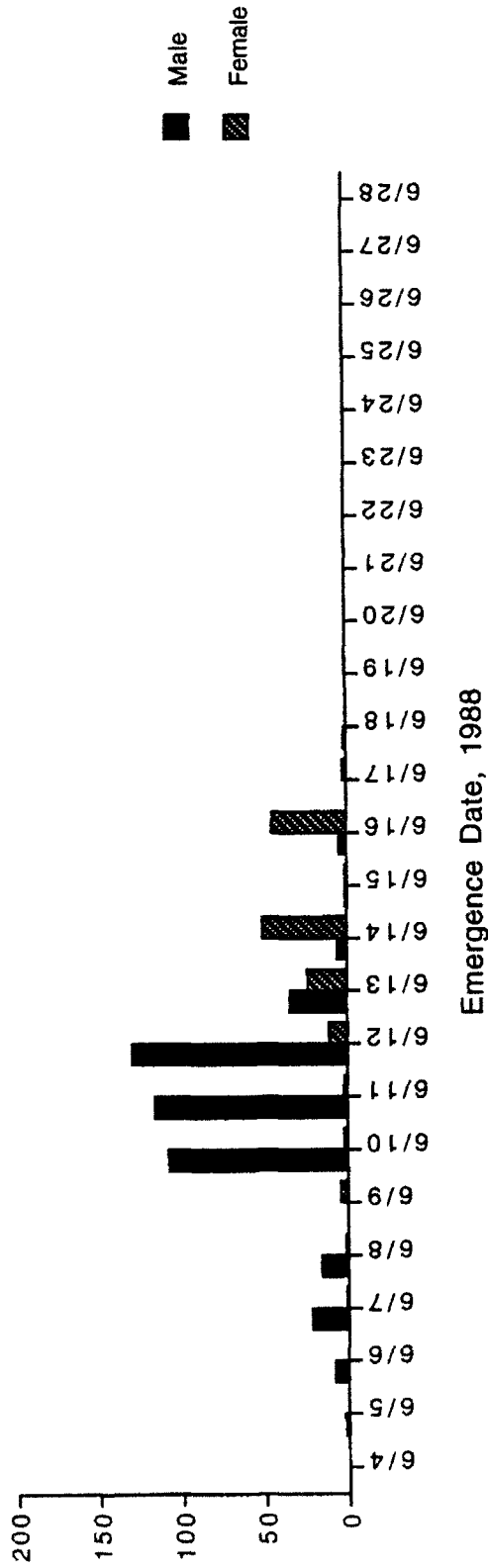
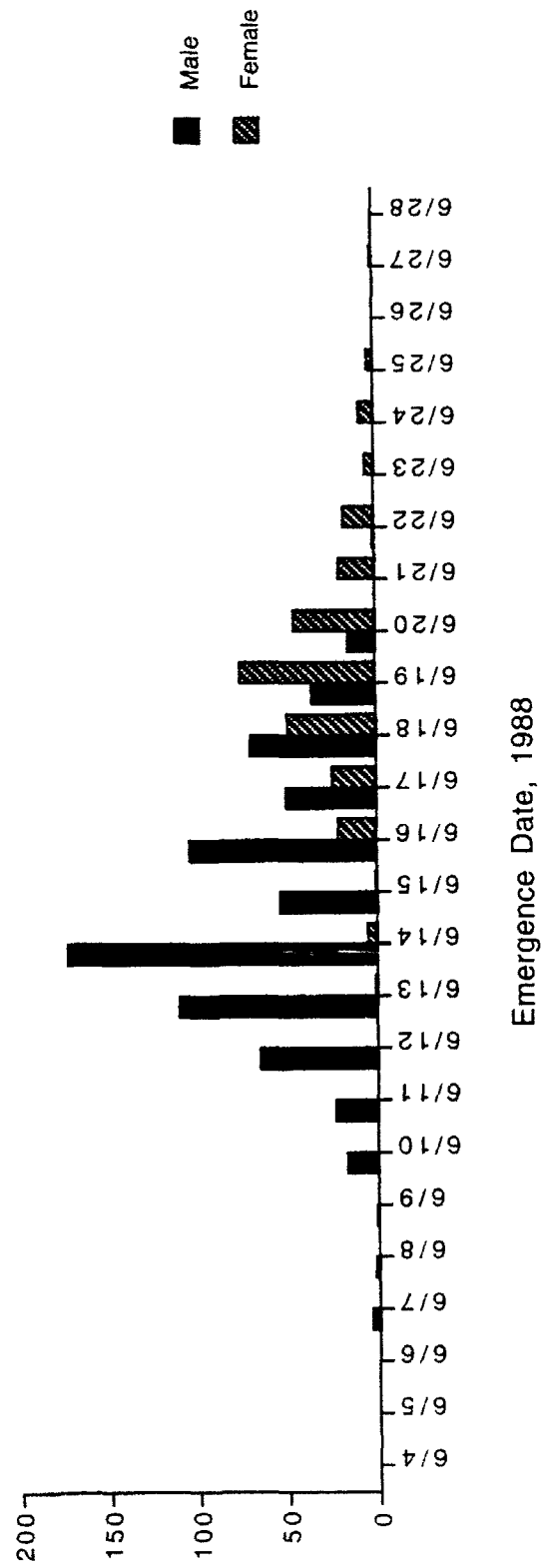
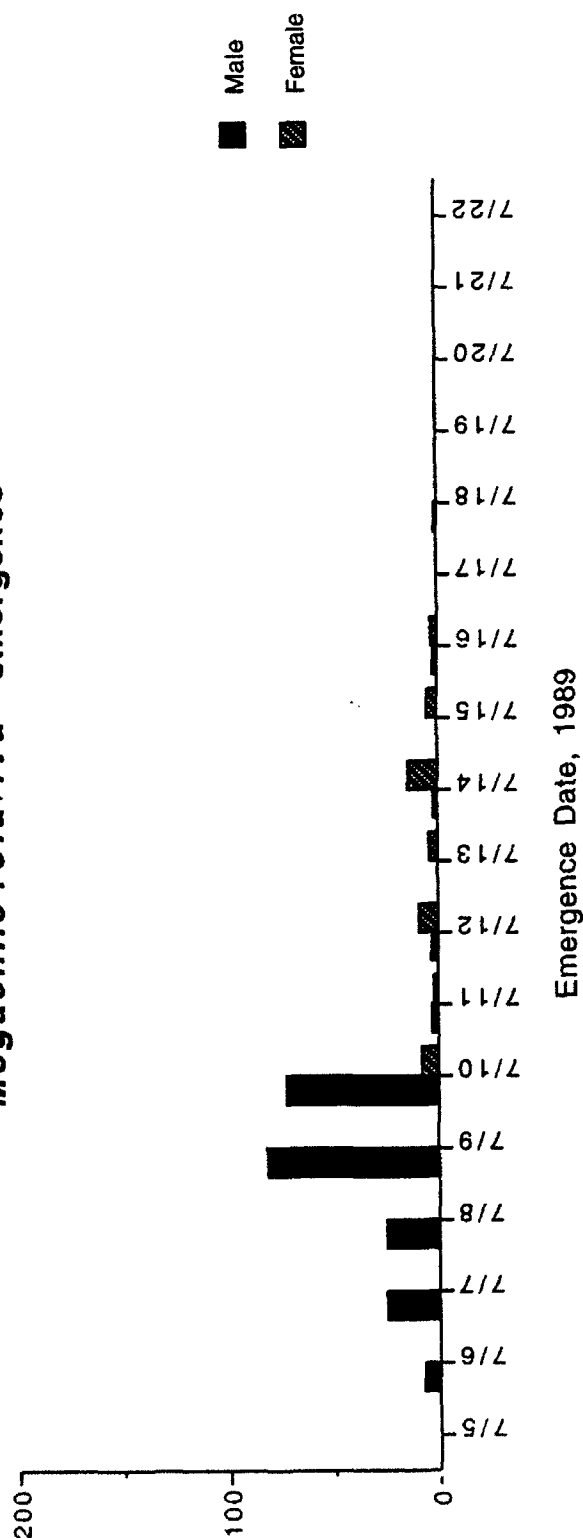
Megachile relativa emergence*Megachile inermis* emergence

FIGURE 29. Phenology of emergence, 1987 nests emerging in 1988.

Megachile relativa emergence



Megachile inermis emergence

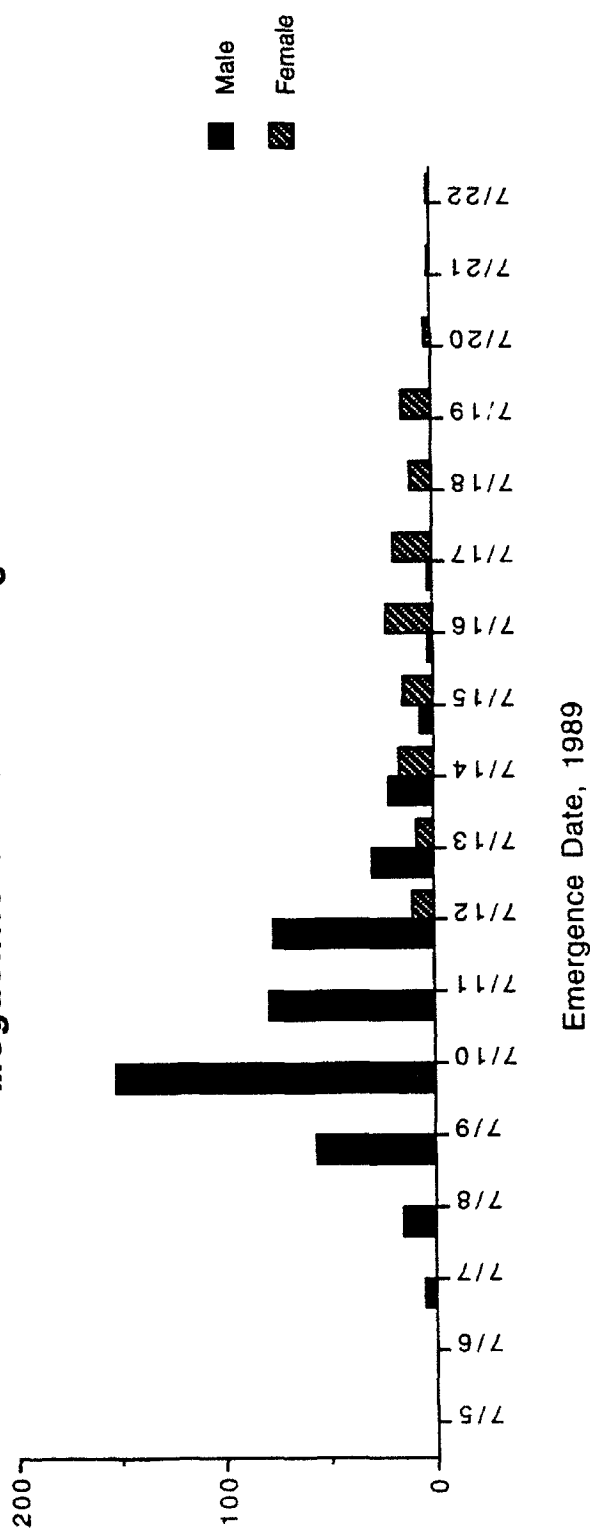


FIGURE 30. Phenology of emergence, 1988 nests emerging in 1989.

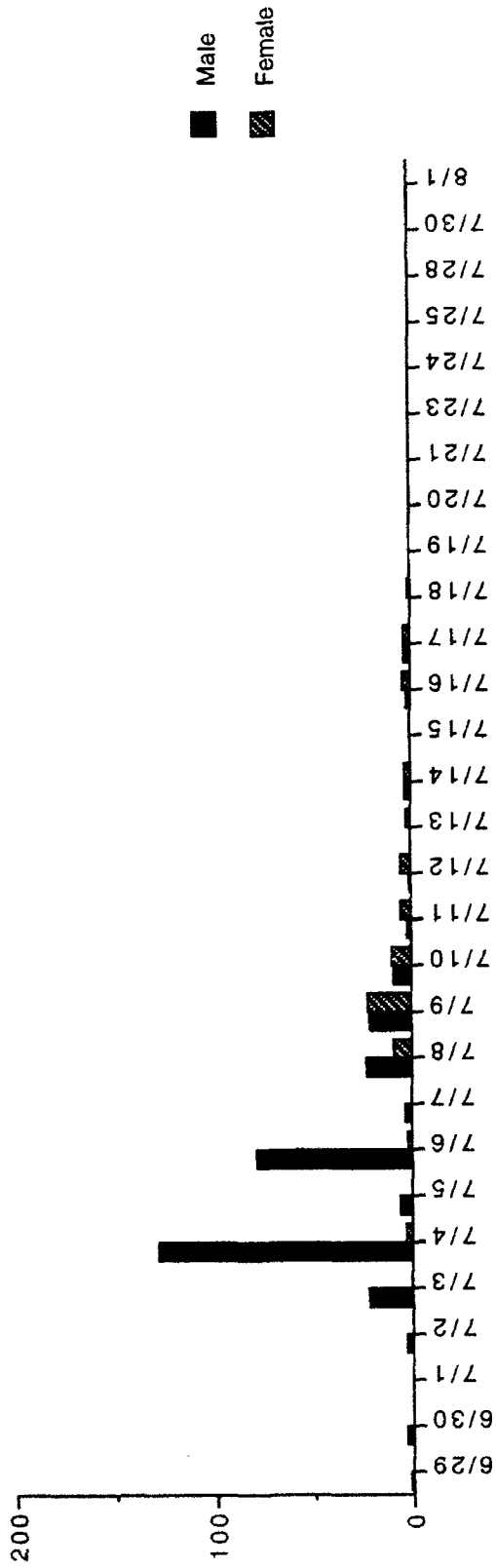
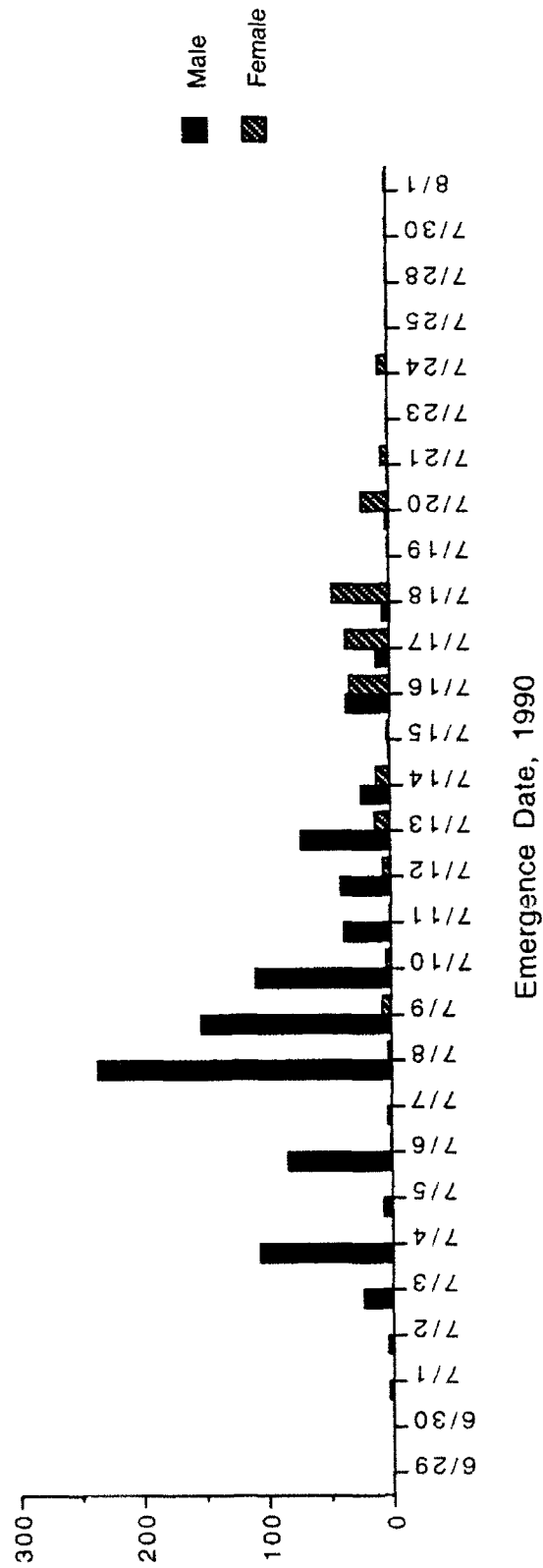
Megachile relativa emergence*Megachile inermis* emergence

FIGURE 31. Phenology of emergence, 1989 nests emerging in 1990.

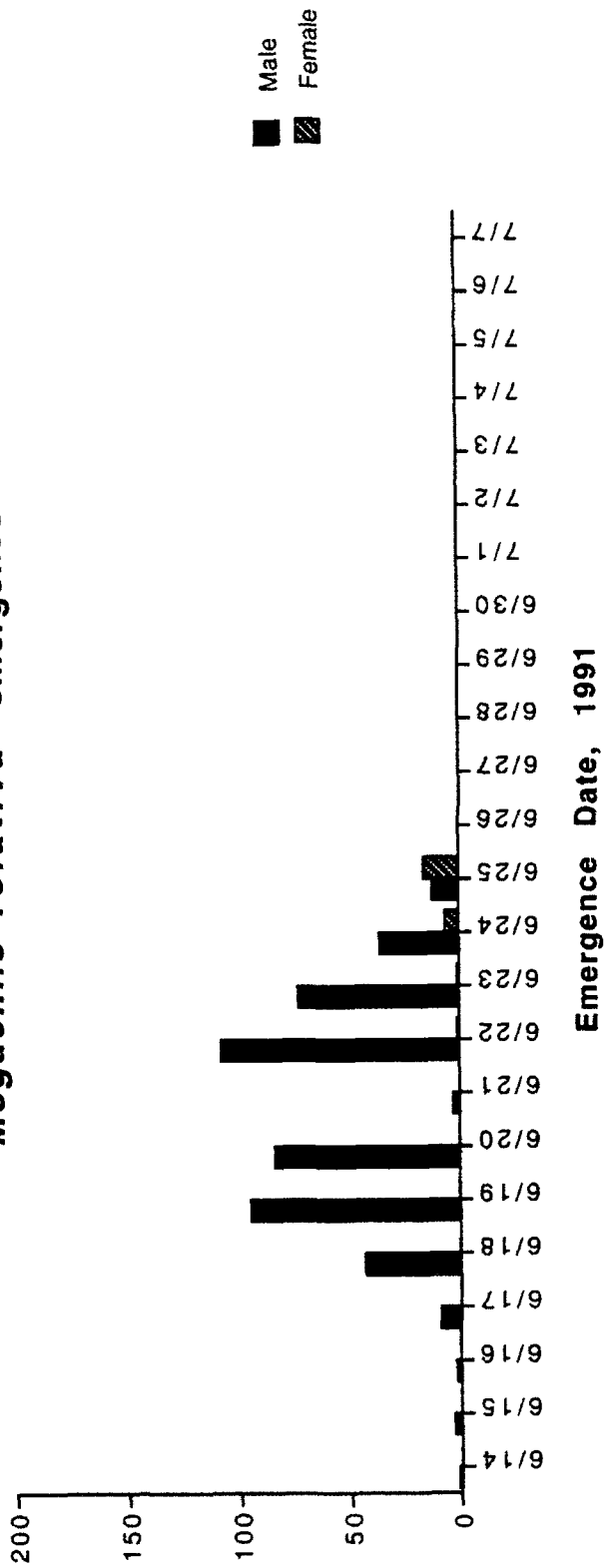
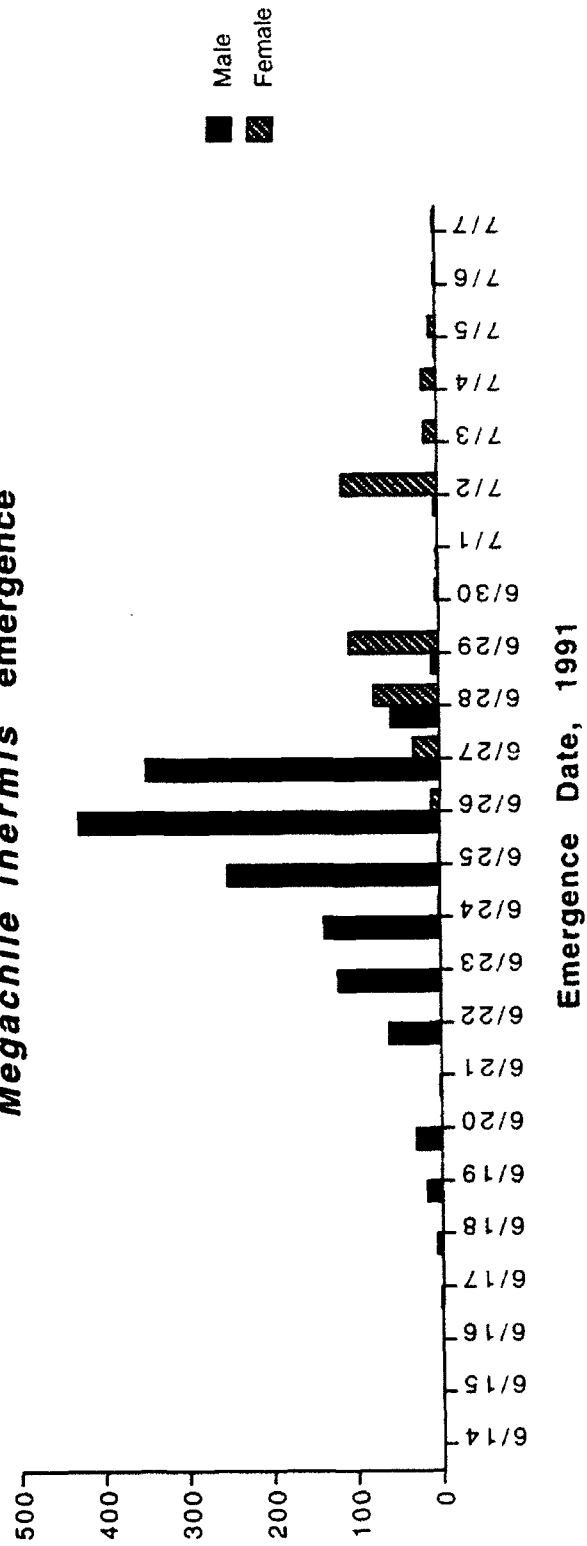
Megachile relativa emergence*Megachile inermis* emergence

FIGURE 32. Phenology of emergence, 1990 nests emerging in 1991.

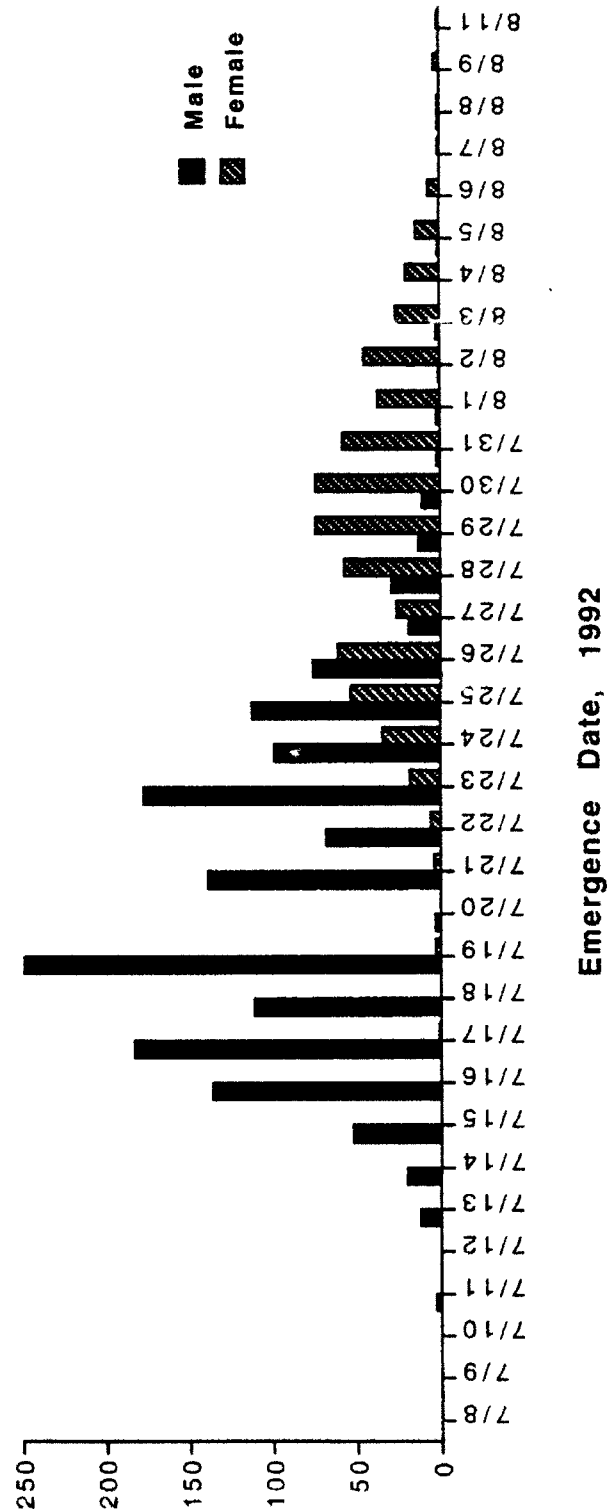
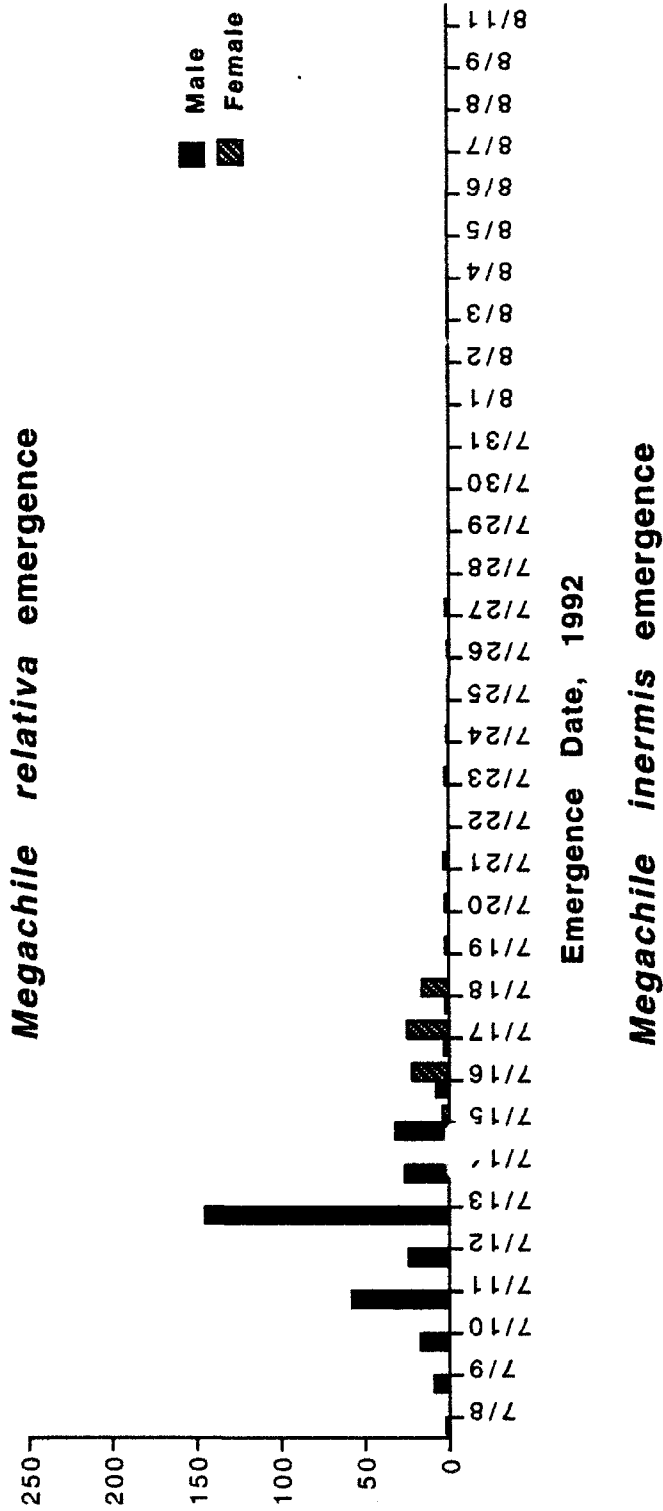


FIGURE 33. Phenology of emergence, 1991 nests emerging in 1992.

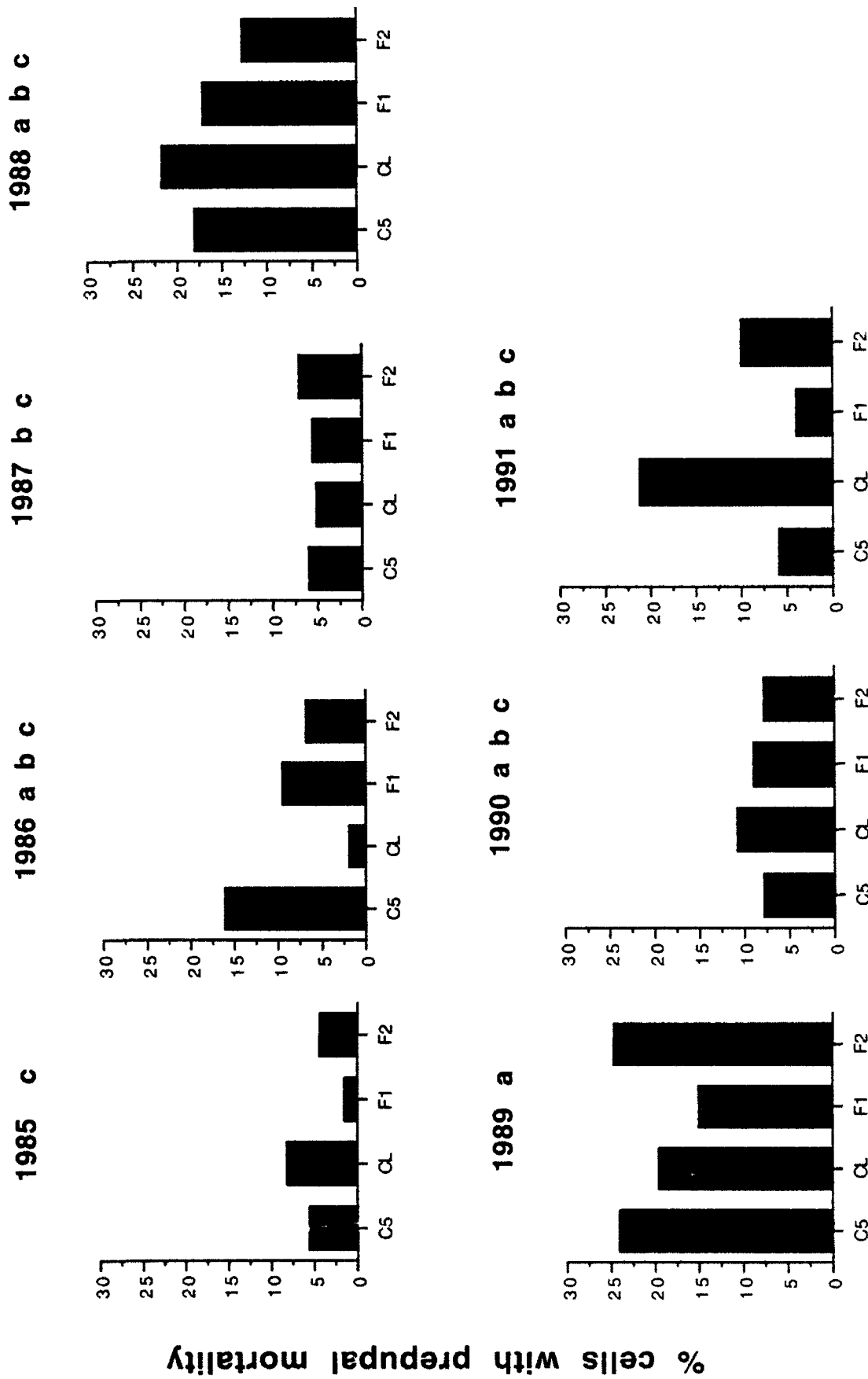


FIGURE 34. Percent of cells with prepupal mortality by year and site, *M. relativa*. Years followed by the same letter are not significantly different.

TABLE 30: CATMOD analysis of cells with prepupal mortality vs. cells with pupae and adults for M. relativa, 1987 - 1991.

PROPORTION OF CELLS WITH PREPUPAL MORTALITY

Source of variation	df	Chi-Square	Prob.
Intercept	1	449.35	0.0000***
Exp	1	2.72	0.0988
Site [Exp]	2	4.02	0.1340
Antenna	1	22.48	0.0000***
Year [Antenna]	3	91.67	0.0000***
Exp * Antenna	1	0.48	0.4877
Residual	11	18.46	0.0715

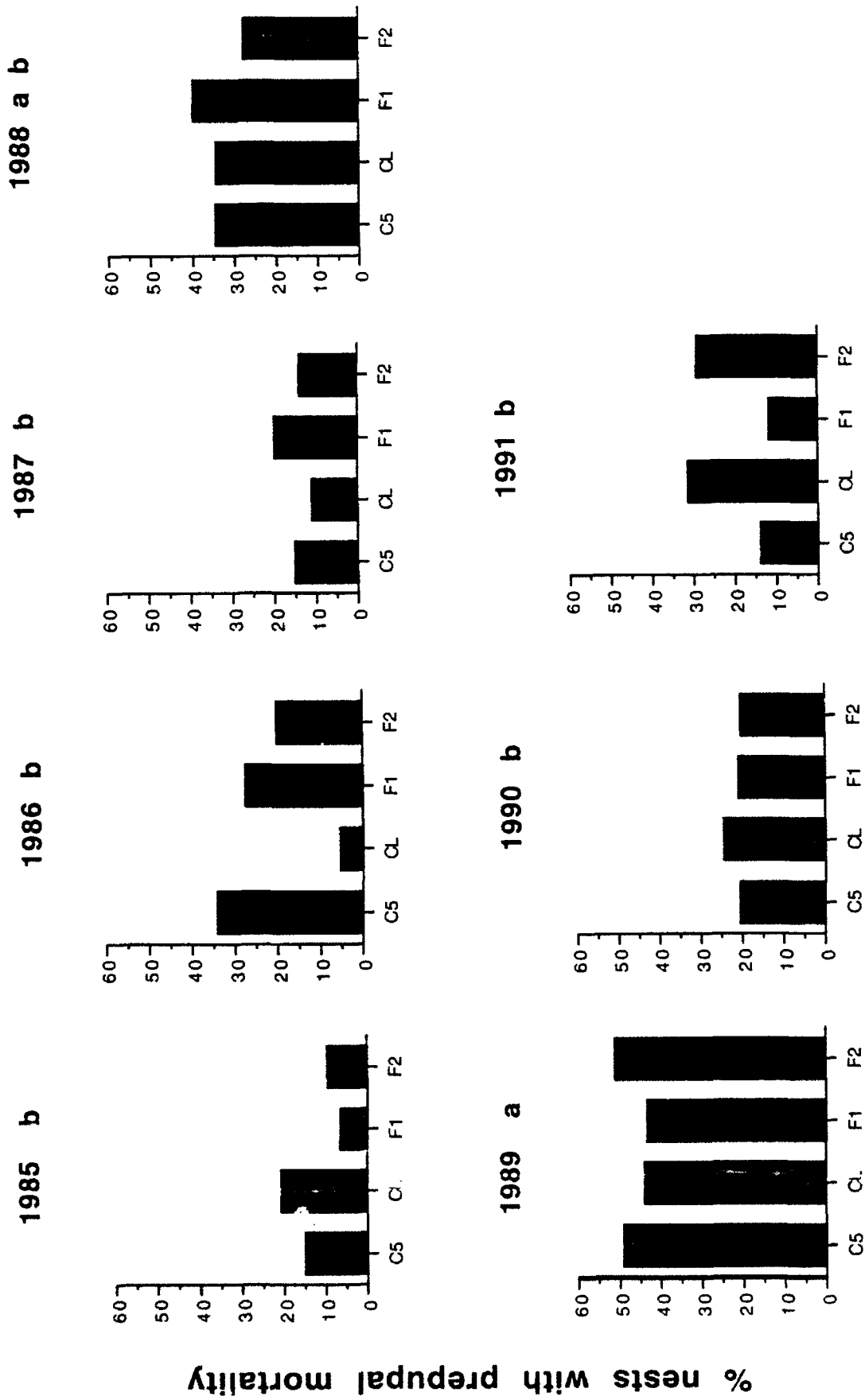


FIGURE 35. Percent of nests with prepupal mortality by year and site, *M. relativa*. Years followed by the same letter are not significantly different.

TABLE 31: CATMOD analysis of nests with prepupal mortality vs. nests with pupae and adults for M. relativa, 1987 - 1991.

PROPORTION OF NESTS WITH PREPUPAL MORTALITY

Source of variation	df	Chi-Square	Prob.
Intercept	1	76.74	0.0000***
Exp	1	0.26	0.6075
Site [Exp]	2	0.20	0.9042
Antenna	1	9.98	0.0016*
Year [Antenna]	3	54.90	0.0000***
Exp * Antenna	1	0.31	0.5754
Residual	11	9.97	0.5328

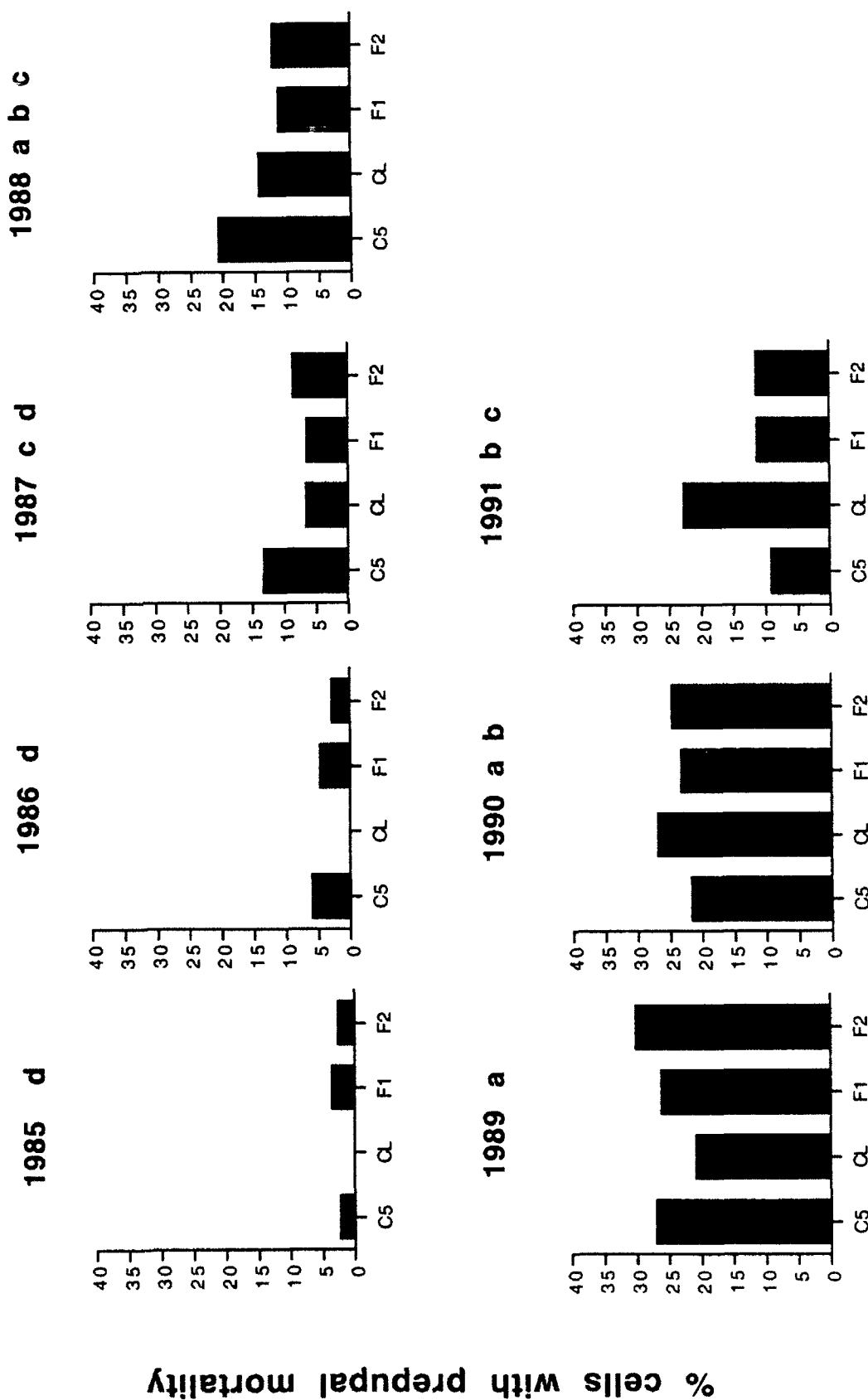


FIGURE 36. Percent of cells with prepupal mortality by year and site, *M. inermis*. Years followed by the same letter are not significantly different.

TABLE 32: CATMOD analysis of cells with prepupal mortality vs. cells with pupae and adults for M. inermis, 1987 - 1991.

PROPORTION OF CELLS WITH PREPUPAL MORTALITY

Source of variation	df	Chi-Square	Prob.
Intercept	1	401.93	0.0000***
Exp	1	11.51	0.0007**
Site [Exp]	2	8.73	0.0127*
Antenna	1	16.43	0.0001***
Year [Antenna]	3	182.55	0.0000***
Exp * Antenna	1	8.85	0.0029*
Residual	11	24.02	0.0127*

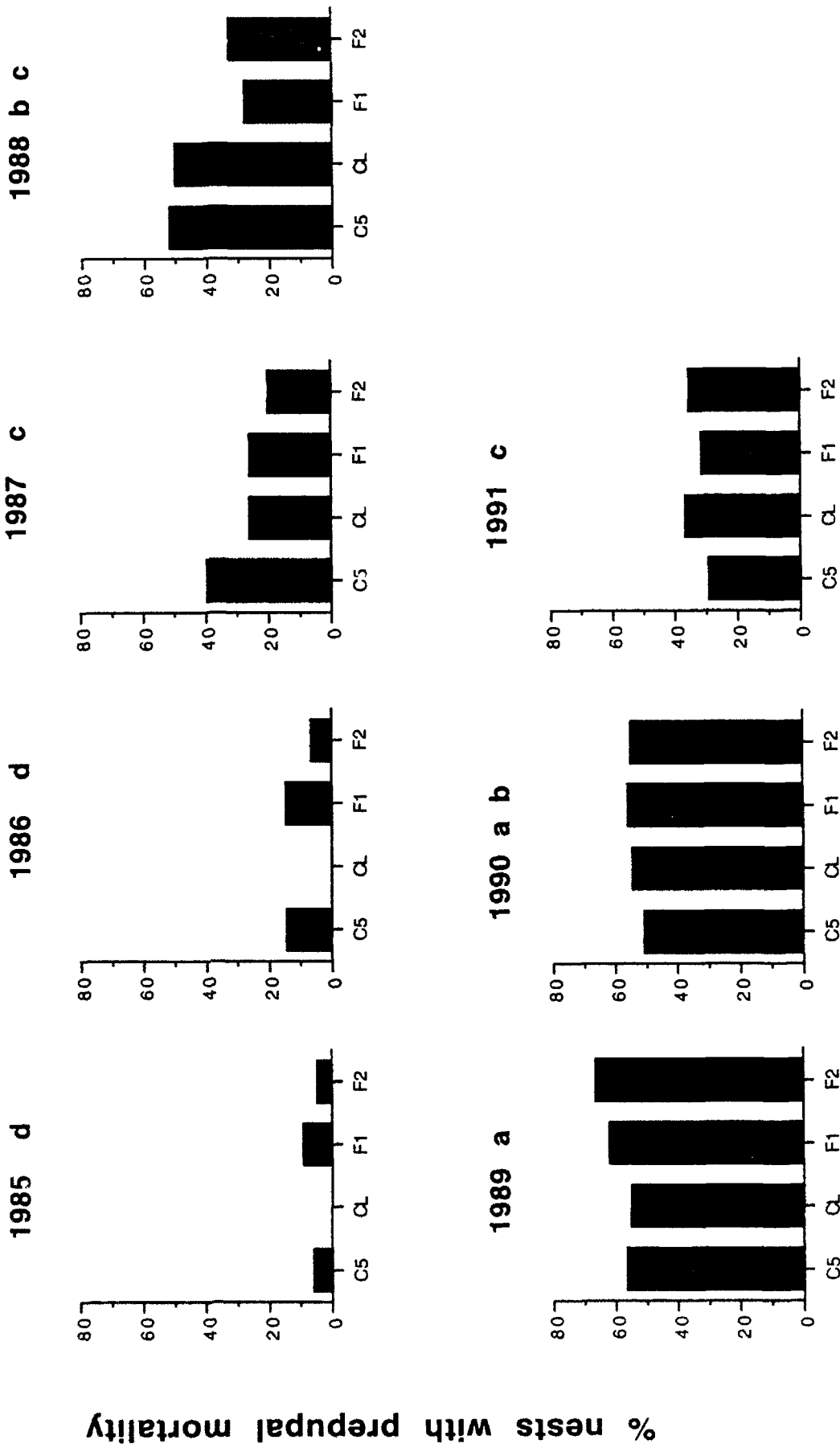


FIGURE 37. Percent of nests with prepupal mortality by year and site, *M. inermis*. Years followed by the same letter are not significantly different.

TABLE 33: CATMOD analysis of nests with prepupal mortality vs. nests with pupae and adults for M. inermis, 1987 - 1991.

PROPORTION OF NESTS WITH PREPUPAL MORTALITY

Source of variation	df	Chi-Square	Prob.
Intercept	1	1.83	0.1761
Exp	1	3.30	0.0694
Site [Exp]	2	0.37	0.8294
Antenna	1	8.25	0.0041*
Year [Antenna]	3	99.58	0.0000**
Exp * Antenna	1	10.90	0.0010**
Residual	11	5.66	0.8951

Overwintering Mortality - *Megachile inermis*

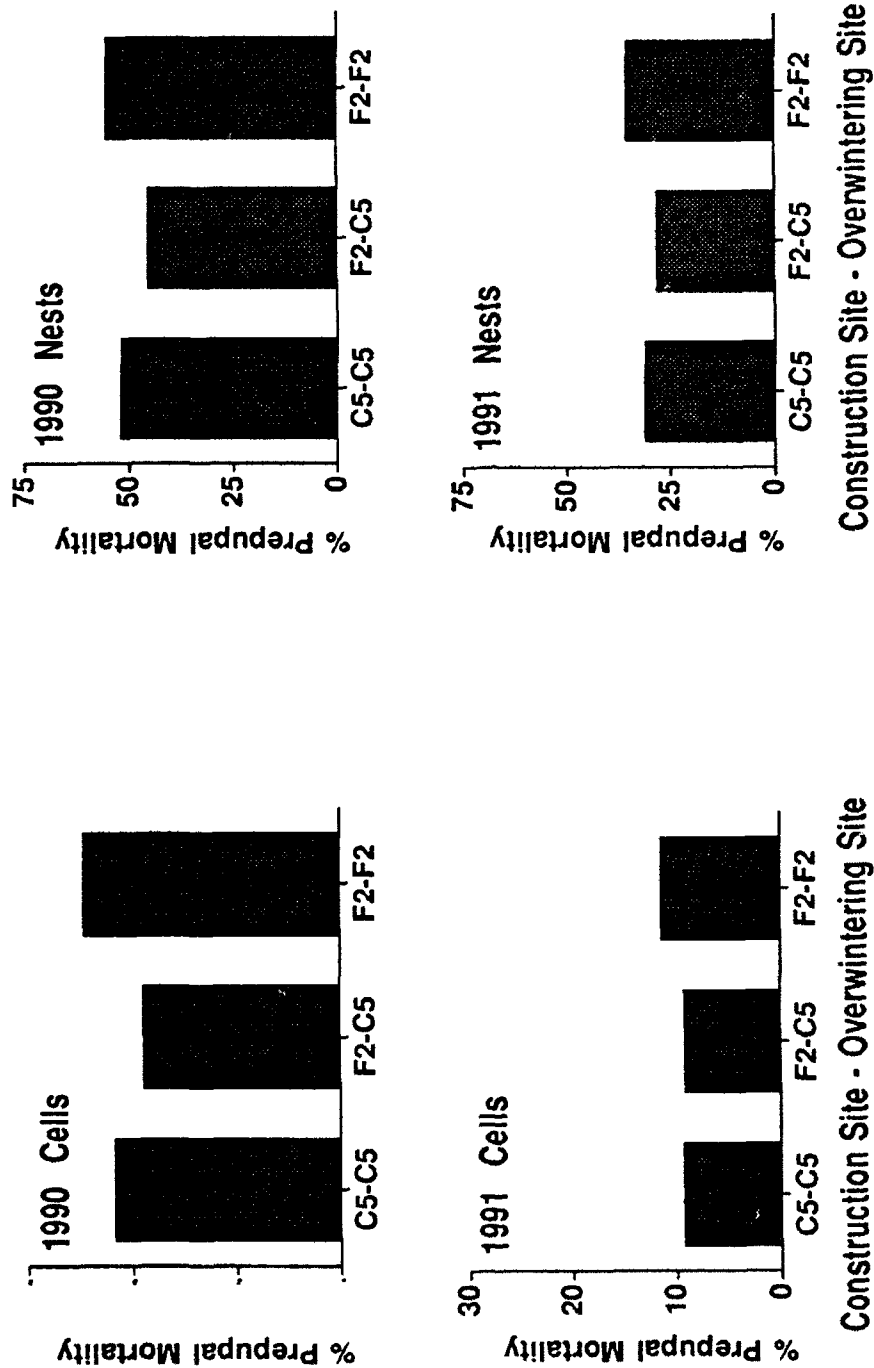


FIGURE 38. Percent prepupal mortality of 1990 and 1991 *M. inermis* from nests constructed at the C5 or F2 site, and overwintered at the C5 or F2 site.

TABLE 34: CATMOD analysis of prepupal mortality of 1990 and 1991 M. inermis cells and nests constructed and overwintered at C5 or F2.

PREPUPAL MORTALITY OF M. INERMIS CELLS

Source of variation	df	Chi.Square	Prob.
Intercept	1	1355.93	0.0000***
Year	1	99.20	0.0000***
Overwintering site (C5 or F2)	1	6.64	0.0100*
Construction site [Ow site] (C5 or F2)	1	0.81	0.3669
Year * Ow Site	1	0.01	0.9247
Residual	1	0.35	0.5538

PREPUPAL MORTALITY OF M. INERMIS NESTS

Source of variation	df	Chi.Square	Prob.
Intercept	1	26.73	0.0000***
Year	1	35.10	0.0000***
Overwintering site (C5 or F2)	1	4.79	0.0286*
Construction site [Ow Site] (C5 or F2)	1	1.03	0.3093
Year * Ow Site	1	0.00	0.9927
Residual	1	0.16	0.6875

ELF COMMUNICATION SYSTEM ECOLOGICAL MONITORING PROGRAM

SMALL VERTEBRATES: THE MICHIGAN STUDY SITE
TASKS 5.6, SMALL MAMMALS, AND 5.12A, NESTING BIRDS

ANNUAL REPORT: 1992

Subcontract No.: E06595-88-C-006

Subcontracted to:

THE BOARD OF TRUSTEES, MICHIGAN STATE UNIVERSITY

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ABSTRACT

The small mammal and nesting bird biological studies in the western Upper Peninsula of Michigan for the year 1992 are reported. Previous years' data include base-line data from 1983-1986 and data collected during partial antenna testing from 1987, through 1989, and full operation in 1990 through 1992.

Data on tree swallow fecundity, survival and growth were analyzed according to antenna operation, plot (test or control) and across years. There were no differences found due antenna operation or due to location plots for any of the following variables: clutch size, distribution of clutch size, likelihood to hatch, hatch rate, likelihood to fledge, number fledging, growth rates (both nonlinear growth constants and linear rates) and maximum size and age at maximum size of tree swallow nestlings for body mass, leg length (tarsus), arm length (ulna), wing length, age at eye opening and feather eruption. These variables show significant year effects due to weather and also significant effects due to the nest they are reared in.

A nestling exchange experiment was instituted in 1990 and repeated in 1991 where randomly selected nestlings were transferred to other nests within and among plots at hatching. Their subsequent growth was monitored and compared in an Analysis of Variance. No effect of the transfer or degree of exposure (as egg or as nestling or both) was detected for either year. A significant year effect was found independent of plot or exchange procedure.

An Analysis of Covariance using insect biomass as the covariate failed to help explain the small differences between plots observed for fecundity measures in some years.

Growth rates, age of incisor eruption and eye opening of young deermice were similar between test and control plots and antenna operation periods. A significant effect of the mother on her nestling's growth and the year was found, paralleling the situation in tree swallows.

The tree swallow homing study was modified to test for the effect of the release site on homing. This was done to see if earlier findings of better

return rates and lower return times by birds from the test site was due to the characteristics of the release site. This year we released test birds from the control release site. We found greater numbers of displaced birds returned to test than control plots, confirming a pattern seen in other years. The time required to return to the plot was also less for test than control birds confirming earlier findings from all years. We conclusively reject the difference as being due to the release site. The significant differences between the performance of test and control birds still stands, although these differences are not due to antenna operation. Significant differences between test and control plots have been observed every year, both prior to the antenna activation and during full antenna operation.

Small mammal homing studies indicated no difference in frequency of return for chipmunks or deermice in 1991. We have obtained mixed results over the years, leading us to conclude there is little evidence for an effect due to ELF electromagnetic radiation.

Developmental abnormalities were not different in number on test and control plots in 1992. Egg weight and measured egg volume were also not different among plots. Both variables showed a significant year effect.

Maximum aerobic metabolism was similar on plots and antenna operation period for deermice. Analysis of covariance using body weight as the covariate did not modify these findings. Chickadees showed no effect due to antenna operation, but did reveal a significant plot effect. However, the interaction between antenna operation and plot was not significant which eliminates ELF electromagnetic radiation as the cause of the plot effect. We do not know the cause of the plot effect.

SUMMARY FOR LAY AUDIENCE

The 1992 report contains results from the biological studies of small mammals and birds from the time period preceding antenna testing (1986) and the partial antenna testing years of 1987 through 1989 and the full operational strength years of 1990 - 1992. While findings must be considered as incomplete until the end of the project in 1993, each year's data is useful in establishing trends in the aspects of small mammal and nesting bird biology at the study sites.

In all years, nesting tree swallows on both test and control plots laid clutches of similar size with a similar likelihood to hatch and fledge on test and control plots. Mortality of eggs, nestlings and nests have been shown to be higher, lower and equal on test compared when to control plots over the years. Because these results are highly variable and in different directions from year to year, data cannot be grouped within antenna operation periods. Growth and maturation (eye opening, feather appearance) of nestling tree swallows showed no difference on test and control plots or period of antenna operation. Thus, there is no evidence of any effect of the antenna's electromagnetic fields on any of the variables we measured. As in previous years, parental care seems to greatly influence nestling growth and parents differ greatly in their ability to raise their young. There is also an effect observed due to weather that causes growth and other variables some years to differ from others. Even though there are parental care and weather differences, they are similar on test and control plots.

Growth and maturation of deermice showed no difference between test and control plots or antenna operation period. As with the tree swallows, mothers showed large differences in their ability to raise their offspring and the weather has a large effect on the young.

Homing studies of tree swallows continued to show higher rates of return and faster return times for birds from test plots. In 1991, we released test plot birds at the same site used for the control plot birds. This was done to control for a potential release site difference. Still, overall times to return were shorter for test plot birds compared to those from control plots, and return rates were better for test birds. These findings eliminate the release site as a factor causing the observed difference. In addition, the differences between test and control plots have been consistent regardless of antenna operational status. This eliminates the antenna as being a causal factor. It is not known at this time what factor(s) are causing the differences we see.

Chipmunks and deermice returned to their home ranges at similar rates on test and control plots in 1992. For chipmunks, we have never detected any difference in homing ability among test and control plot animals. We have

observed inconsistent results for deermice in prior years, with some years indicating better return rates for animals displaced on test plots and other years for control plots. These inconsistencies lead us to believe there is no antenna effect on homing.

Abnormalities of tree swallow embryos showed no difference in frequency between test and control plots. Egg weight and volume were also not difference between test and control plots or antenna operation periods. A significant effect was found for the year, probably due to weather, and to the parent.

Maximal metabolism of deermice and chickadees showed no difference for test and control plots or for period of antenna operation. Chickadees do show a gradual lowering of their maximal metabolism over time, but we cannot attribute this to the antenna's electromagnetic radiation.

In summary, no effects due to the antenna's electromagnetic radiation has been found for the many variables we are studying, except for homing behavior in tree swallows. We can not at this time be certain that the better performance observed by test plot birds is due to the antenna and not other factors associated with the plot or route of travel.

PREFACE

This report begins with an extensive statement of the rationale for the studies proposed (see next section, titled "Rationale for Proposed Studies"). Then a section is provided on the overall research design and research facilities. Individual elements of the work are then described in detail in a series of subsequent sections. Each of the sections on individual work elements consists of three parts: (1) a brief restatement of the purpose (rationale) for the work, (2) a detailed description of research methods, and (3) a presentation of representative results gathered during prior years. The presentations of results include discussions of statistical sufficiency, including projections of the sample sizes required to discriminate between test and control plots in future years.

RATIONALE OF STUDIES

Dozens of species of small birds and mammals are resident near the ELF Communication System, in the upper peninsula of Michigan, and the operation of the Communication System could in principle affect any of them in any of countless ways. Even with virtually unlimited resources, it would be impossible to monitor individually all ecologically important aspects of all species for possible effects of the Communication System. Accordingly, we have had to exercise informed judgment in selecting variables for study. In this process, we have been guided by one overriding goal.

Our major goal has been to focus much of our effort on attributes of *individual* animals that are particularly likely to be susceptible to perturbation by the ELF Communication System. The reason for this focus is that laboratory research indicates that if the ELF Communication System is to have effects on birds or mammals, the effects will likely be small, and thus a statistically robust experimental design will be required to detect them (AIBS 1985). Large numbers of independent measures can be readily obtained on *individual* attributes, thus facilitating statistical detection of even small effects that the ELF Communication System might have.

In our studies of attributes of individual birds and mammals, we emphasize ecologically significant variables that are especially likely to be susceptible to perturbation. Reproduction and development, for example, receive particular attention because they not only are demographically important but also are more likely to be sensitive to adverse environmental changes than many other animal properties (e.g. Goodposture 1955, Koskimes 1950, Kluijver 1951, Krebs 1971, Lack 1954, 1966, Nice 1954, Perrins 1965, Perry and Rowlands 1973). Behavior is studied in depth because it is sometimes modified readily and such modifications can have major repercussions on the lives of individuals and populations (e.g. Cohen et al. 1980, Green 1979, Morse 1980, O'Connor 1978, Slobodkin 1968).

In the following paragraphs we describe in detail the rationale for each aspect of our work on individual attributes. This work is concentrated on

four particularly abundant species. The species have been carefully selected with a view to maximizing their ecological and taxonomic diversity, so as to maximize the probability of detecting whatever diverse effects the ELF Communication System may have. The four are the tree swallow (*Tachycineta bicolor*), the woodland deer mouse (*Peromyscus maniculatus gracilis*), the black-capped chickadee (*Parus atricapillus*) and the eastern chipmunk (*Tamias striatus*). To facilitate readability in the remainder of the report, they will be referred to simply as the "tree swallow", "deer mouse", "chickadee" and "chipmunk", respectively.

Behavioral Studies

In view of the established sensitivity of certain types of orientational behavior to alteration by the ELF fields (e.g. Graue 1974, Keeton et al. 1974, Larkin and Sutherland 1977, Southern 1969, 1971, 1972a, 1972b, 1973, 1974, 1975, 1976), orientation and homing in the tree swallow, deer mouse, chipmunk, and certain other mammals are being tested to see if they are affected by the ELF Communication System. Specifically, the ability of animals to return to their home-range or territory after displacement is being assessed. We know that animals are able to find food (Krebs 1971, Royama 1966) and escape predators (Metzgar 1967, Watson 1964) more effectively in their home-range or territory than in less familiar areas. Thus, any disturbance of their ability to return to their home-range or territory after wandering afar could decrease their probability of survival.

The attentive behavior of parental tree swallows and deer mice is being assessed by monitoring visits to the nest containing eggs and young. Disturbance of attentive behavior by the ELF Communication System, if it occurred, could impair development of eggs or nestlings inasmuch as the latter are dependent on parents for both food and warmth (e.g. Balen and Cove 1972, Hill 1972b).

Reproduction, Growth, and Development

The frequency and type of prenatal developmental abnormalities are examined in tree swallows. Mammals are not studied in this respect because reproductive females would have to be killed to examine fetuses, and such deaths could have serious, adverse effects on population demographics. Prenatal developmental stages are especially likely to be susceptible to perturbation (Axelsson 1954). Developing avian embryos have two major periods of sensitivity (Hamilton 1952) which occur during the first 4 days following the onset of incubation and the period just prior to hatching. A majority of the spontaneously occurring developmental abnormalities manifest themselves during these two periods (Riddle 1930, Hutt and Pilkey 1930, Hutt and Greenwood 1929, Hutt and Crew 1929, Landauer 1943, Martin and Insko 1935, Hamilton 1952). During these periods, the embryos are sensitive to changes in naturally occurring environmental agents such as temperature, humidity, CO₂, and O₂ (Alsop 1918, Babott 1937, Pembrey et al. 1894, Romanoff et al. 1938, Taylor et al. 1933). Additional teratological agents include vitamins and their antagonists (Cravens 1952), hormones (Zwilling 1956), alcohol and ether (Stockard 1914), metal ions (Ridgeway and Karnofsky 1952), narcotics (Reese 1912), various forms of radiation (Windle 1893, 1895, Gilman and Baetjer 1904, Hinrichs 1927, and Dixon 1952) and physical jarring (Stiles and Watterson 1937). Since the onset of this investigation, effects of ELF radiation on chick development have been reported (Delgado et al. 1982, Ubeda et al. 1983, Juutilainen and Sali 1986, Juutilainen et al. 1986). There is, at present, no evidence to demonstrate that electric and magnetic fields of the magnitude generated by the ELF Communication System are capable of directly causing embryonic or fetal developmental defects. However, indirect effects are possible. Should the incubation behavior of parent birds be disturbed by the ELF Communication System, developing eggs might suffer developmental abnormalities by virtue of experiencing abnormal reductions or fluctuations in temperature (Zwilling 1956, Hamilton 1965).

We monitor aspects of fecundity in both tree swallows and deermice. In the birds, we count the number of eggs produced per female and the number of

viable eggs and young per clutch. In the mice we monitor numbers of young per litter. Fecundity is an important variable to study not only because it is demographically significant but also because it reflects on a number of variables that could, in principle, be affected by the ELF Communication System. Alteration of male or female reproductive physiology could affect fecundity. Further, any serious disturbances of prenatal development in mammals or birds would likely be reflected in a decrease in fecundity inasmuch as abnormal embryos frequently fail to be born (i.e., they are resorbed in utero or fail to hatch) or are eaten or discarded by the parents soon after birth.

Postnatal mortality and the growth and development of nestling tree swallows and deermice are also followed. Any effects that the Communication System might exert on the young themselves could be reflected in altered rates of mortality, growth, or development. Alternatively, disturbances of parental attentive behavior could be influential because the rates of mortality, growth, and development of nestlings are dependent on the extent to which parents provide food and warmth (Hill 1972b). The size of nestlings at the time of weaning or fledging is of particular interest because when young become independent of their parents, they must become substantially self-sufficient and their maturity can affect their likelihood of survival. Evidence exists that young birds that are of relatively small size at fledging are significantly less likely to survive than ones that grow to larger size while in the nest (Lack 1966, Murphy 1978, Perrins 1965).

Maximal Aerobic Metabolism

In the region of the ELF Communication System, low temperatures make winter the most physiologically stressful time of year, at least for animals such as chickadees that live wholly or predominantly above the snow. We study physiological variables that affect the ability of chickadees and small mammals to cope with the severity of the winter climate. Deficits in the physiological ability to cope would be expected to decrease the probability of survival to the next reproductive season.

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Birds and mammals keep warm in cold environments by producing heat metabolically to offset heat losses. The extent to which they can keep their body temperature above air temperature depends on how rapidly they can produce heat. In other words, the lowest air temperature at which they can maintain their usual body temperature is a function of their maximal rate of aerobic metabolism (= heat production) (Hart 1957). In view of these principles, we measure the maximal rate of aerobic metabolism of chickadees and deermice during winter. This peak rate of heat production is informative not only because it determines the lowest air temperature at which thermoregulation is possible but also because it likely provides an index of metabolic endurance. The higher an animal's maximal rate of heat production is, the longer the animal will be able to maintain any particular submaximal rate of heat production (Astrand and Rodahl 1977, Wickler 1980). Endurance is important because low air temperatures demanding high heat production can persist for long periods of time.

Beyond its immediate significance for survival in a cold climate, the maximal rate of aerobic metabolism is a valuable variable to measure because it provides an index of physiological health. In fact, peak aerobic metabolism is widely used as such an index in studies of humans. In their classic *Textbook of Work Physiology*, Astrand and Rodahl (1977) state that "the maximal oxygen uptake is probably the best laboratory measure of a person's physical fitness" if by fitness we mean the capacity of the individual for prolonged heavy work. Brooks and Fahey (1984), in the best of the recent texts on human exercise physiology, state that the maximal aerobic metabolism is "a good measure of fitness for life in contemporary society". Just as peak aerobic metabolism is used as an index of fitness for humans, it can also be so used in studies of animals. A deficit in the peak metabolism among individuals living near the ELF antenna would indicate that some attribute of the all-important systems involved in oxygen supply and use has been adversely affected by the ELF electromagnetic fields. Additional tests would then be required to determine the particular attribute(s) affected. The ability of the respiratory system to provide oxygen, the ability of the circulatory

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system to transport oxygen and nutrients to metabolically active tissues, the ability of storage tissues (e.g., adipose tissue) to mobilize stored nutrients, and the enzymatic competence of metabolically active tissues to catabolize nutrients are among the variables that influence an animal's peak rate of aerobic metabolism (Wang 1978). In human studies, peak aerobic metabolism is usually elicited by having individuals run on a treadmill. We elicit peaks by exposing animals to cold, in part because the method is technically simpler than treadmill running (given that animals require extensive training to use a treadmill successfully) and in part because the cold-induced peak is of immediate relevance to understanding winter ecology.

OVERALL RESEARCH DESIGN AND SUPPORT FACILITIES

To detect possible effects of the ELF Communication System, we compare animal attributes on test plots (test sites) with those on paired, spatially separated control plots (control sites).

Test plots, as specified in the original IITRI Request for Proposals, are areas close enough to the Communication System that electric and magnetic fields attributable to the System, and measured in the soil near the earth's surface, will approximate 0.07 volt/meter and 0.03 Gauss, respectively. Furthermore, electric and magnetic fields attributable to ELF sources other than the System are to be at least an order of magnitude lower than those attributable to the System.

Control plots, according to the original Request for Proposals, are areas sufficiently distant from the Communication System that electric and magnetic fields attributable to the System, measured in the soil near the earth's surface, are at least an order of magnitude, and preferably two orders of magnitude, below those at paired test plots. Furthermore, electric and magnetic fields in the air and earth attributable to ELF sources other than the System (especially 60 Hz sources) are not to differ by more than an order of magnitude between the control plots and their paired test plots.

For purposes of experimental design, the test plot(s) used for any particular work element are paired with particular control plot(s). The plots of a pair are matched as closely as possible for vegetation, soil type, drainage, and other such features. By pairing plots in this way, we minimize the likelihood that non-ELF differences between plots will introduce significant confounding effects into our results.

A major strength of our research is the paired plot design. Within a year, we can compare possible ELF effects across plots. The design has an additional strength due to the capability of before and after comparisons for each plot where each plot can be used as its own control through time. We consider three phases of antenna operations: 1) pre-antenna, 1983-1985, 2) antenna testing, 1986-1988, and 3) full antenna operation, 1989-1991.

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Different work elements are carried out on different pairs of plots for several reasons. Specific work elements could interfere with other work if both were carried out on the same populations of animals; areas where we artificially remove animals (e.g., bird embryos), for example, are not used for research on natural populations. Another factor that demands the use of different plot pairs for different work elements is that the various species we study do not all occur in similar habitat types; field habitats are required for the swallows, whereas forests are required for the deer mice.

To minimize potentially confounding differences between test and control plots, sham corridors have been cut through the forests at the control plots. These corridors are clearings of the same width as the corridors cut for installation of the Communication System antenna near test plots. They were cut with similar equipment, and they have been treated similarly after cutting. In brief, the sham corridors are as identical as possible to the antenna corridor except that antenna poles and wires have not been installed in the shams. Areas for animal study on control plots and those for animal study on test plots are located about the same distance from the sham corridors and antenna corridor, respectively.

Study Plots. Plots were established as matched pairs of test and control plots (?) for the various work elements. Test plots were located along the north-south antenna element and control plots were located at varying distances to the west of the antenna (?). The names given to the plots (Table 1) are the standardized ones we use in all our descriptions of experiments and results. The alpha-numeric codes for plots are those used by IITRI.

Modifications in Project Scope and Statistical Sufficiency

The number of study elements was reduced in March, 1989, when budget cuts were made to meet increased wages of non-faculty employees on the project. The wage increases resulted from a labor settlement at Michigan State University. The following research elements were dropped: small mammal community studies, small mammal parental care, and tree swallow incubation. All remaining research elements were continued at full strength.

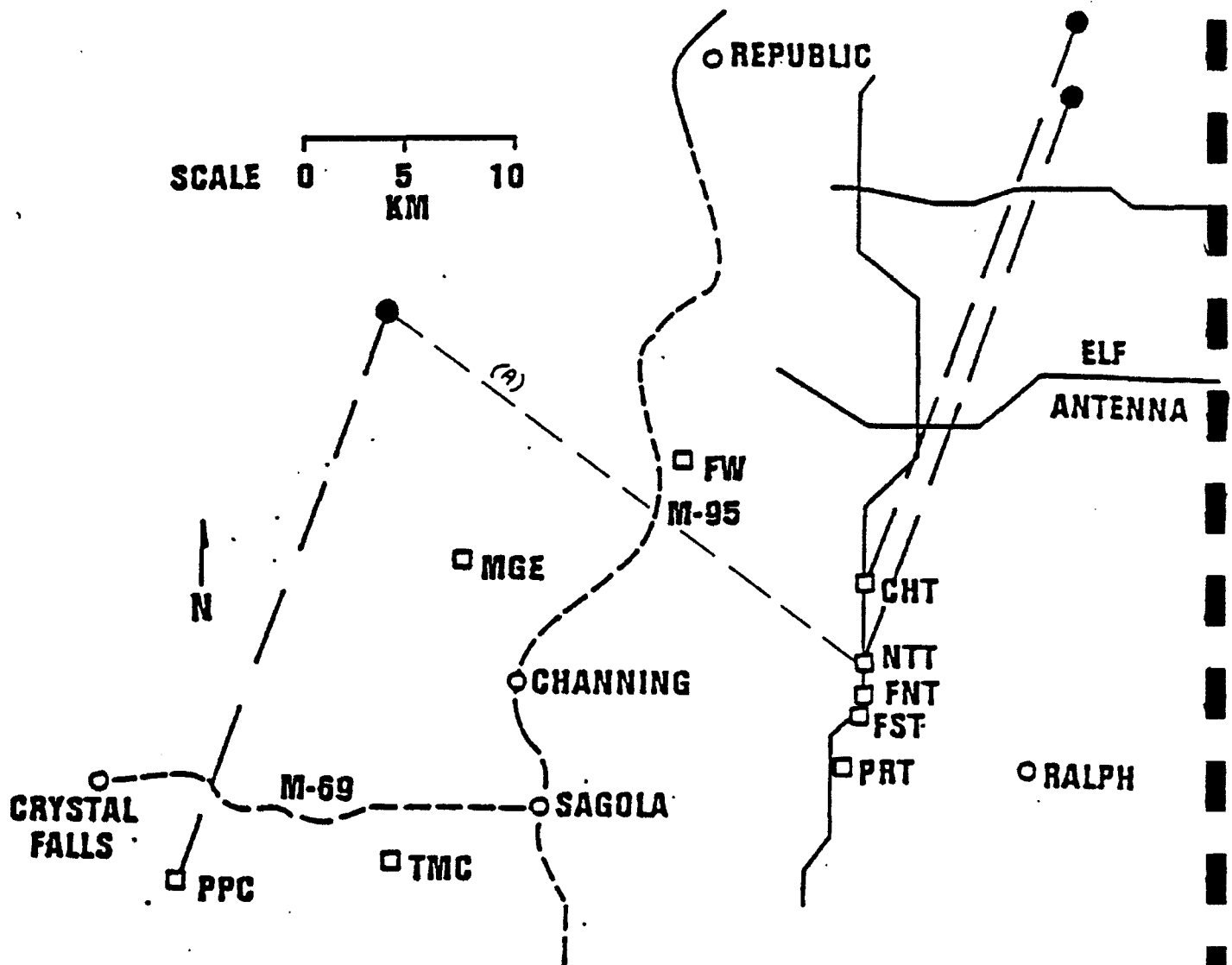


Figure 1. Location of test and control plots in relation to the antenna system. See Table 1 for plot codes

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Table 1. Test-control plot pairings for the various work elements for small mammals and nesting birds. Plot code designations are those used by IITRI.

STUDY ELEMENT	TEST PLOT	CONTROL PLOT
Deermouse Growth & Maturation	PIRLOT ROAD (1T1)	MICHIGAMME NORTH (1C1)
Small Mammal Homing	PIRLOT ROAD (1T1)	MICHIGAMME SOUTH MICHIGAMME NORTH (1C3, 1C1)
Deermouse Winter Physiology	PIRLOT ROAD (1T1)	MICHIGAMME SOUTH (1C1)
Tree Swallow Growth & Maturation	PIRLOT ROAD (1T1)	TACHYGINETA MEADOW (1C6)
Tree Swallow Homing (Home Plots)	CLEVELAND HOMESTEAD (1T2)	PANOLA PLAINS (1C4)
	NORTH TURNER ROAD (1T4)	PANOLA PLAINS (1C4)
(Displacement Plots)	CLEVELAND HOMESTEAD DISPLACEMENT (1D1)	-
	NORTH TURNER DISPLACEMENT (1D2)	-
	-	PANOLA PLAINS DISPLACEMENT (1D3)
Tree Swallow Embryology	-	TACHYGINETA MEADOW (1C6)
	FORD RIVER NORTH (1T5)	PANOLA PLAINS (1C4)
	FORD RIVER SOUTH (1T6)	
Black-capped Chickadee Winter Physiology	PIRLOT ROAD (1T1)	MICHIGAMME NORTH (1C1)

Note: Ford River North and Ford River South plots are small. Therefore they have been designated solely as tree swallow embryology study sites.

We have revised our standards for statistical sufficiency for the

research program based on our years of experience with the various study elements to date. We originally established the standard of statistical sufficiency in our work that we predicted would provide a 90% certainty of detecting a 20% difference between test and control sites at the 5% level of significance. While we can still meet these standards on some of our work, we clearly cannot for others, such as growth of both tree swallows and deermice (see Tables dealing with growth). Variation among nests unrelated to plot is the principle reason. These unexpectedly high variances lead to projections of sample sizes beyond the possible scope of our research. We must therefore relax our standards of statistical sufficiency. We have decided to report the actual level of detectable difference in means achieved in a test and the difference we could detect if we relaxed the level of certainty (power) to 70%. The reader will therefore be able to judge for each test the particular statistical confidence that can be met. Literature values for detectable differences or power are not currently known to us for comparison. It seems that most authors do not report either value. For discontinuous variables, we have used different procedures to determine sample size (see Gill 1978, p. 82). Therefore we do not propose changes in statistical sufficiency since we appear to be able to meet the stricter requirements with these data. Discussion of sample size and power of test are presented with the data for each study element (see below).

Our base of operations for the on-site field and laboratory studies is a large house rented in Crystal Falls, MI (801 Crystal Ave.). The physiology laboratory is installed there. The holding facility for temporary housing of animals used in the physiology experiments is located approximately 3.5 miles south of Crystal Falls, MI in an area with minimal electromagnetic interference. We have a shop for construction and maintenance of field equipment and a large shed for storage of traps, cages, construction materials, and seasonal field equipment. We also have a well established data management system housed there (see below), and living space is provided for employees. We lease and maintain trucks to provide transportation between our base of operations and field research sites in all weather conditions on a

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year-round basis. In addition, we rent a snowmobile to gain access to our more remote sites during winter and spring when traveling the entire distance by truck becomes impossible.

For data management we employ an IMS (now LF Technologies) computer system at the MSU Museum in East Lansing. The system is multi-user and allows storage of data on fixed and removable media. Zenith (AT-class) computers are used at the field laboratory in Crystal Falls. Data transfer and analysis are accomplished using both systems. Field data are collected by NEC PC-8201A portable computers. We have developed software to standardize and error check field data as it is recorded. Collected data are transferred directly into an AT computer at the field laboratory each day. Transferred data are immediately edited and stored on removable and fixed disks for later analysis. Certain data are analyzed as soon as they are collected. This data management design allows us to collect and analyze large amounts of data very efficiently and accurately. In addition, in 1987, we added high speed tape backup systems to aid in recovery of data should either computer system fail, and for archiving the now voluminous data sets for the various study elements. The large sample sizes required in many of our study elements necessitate the careful and accurate data handling the system provides.

Other major equipment is described in connection with individual work elements in the sections that follow.

Measurements on 60 and 76 Hz fields. Engineers from IITRI have measured 60 Hz electric and magnetic field intensities every year starting in 1983 on our test and control plots, and all the pairs we now use adequately meet the standards for field intensities already described. Electric and magnetic fields produced by the antenna system (76 Hz) were measured starting in 1986, when low amperage testing began. Measurements have continued as the antenna has become operational. A summary of the data 1983-1990 is provided in Tables 2 - 9. Details of the results of the field-intensity measurements and the measurement techniques can be found in Enk and Gauger (1985), Brosh et al. (1985 and 1986), and Haradem et al. (1987, 1988, 1989 and 1991). Earlier

discussion of measures and plot pairings are outlined in the 1984 annual report (Beaver et al. 1985, pp. 3-9).

In all years, measures were made in September or October by IITRI personnel on our test and control plots during antenna operation. The distribution of operation hours by month for 1986, 1987 and 1988 for the north-south and east-west antennas were concentrated in the months of June through November in 1986 and 1987. Continuous operation began in 1988, but the antenna was shut down for repairs during most of the months of January, February and March, 1989, during our winter studies. Continuous operation occurred throughout the remainder of 1989 and all of 1990, 1991 and 1992 (Haradem and Gauger 1991, and personal communication). During these years, the amperage of antenna operation varied from 3 to 150 amperes. Schedules of research activities in the spring and summer fell within the times of heaviest antenna operation in all years. Operation of the antenna was conducted on a 33% time rotation schedule in which the east-west antenna was on for 5 min, then the north-south antenna for 5 min, followed by both antennas off for 5 min. The percentage of time the MTF was on varied from 1.8% (1986) to nearly 100% (1990). The antenna was off for repair and maintenance for about 5 hours twice per week in 1990 (Haradem and Gauger 1991).

60 Hz Fields - Background measures. Measurement of background 60 Hz fields on control and test plots began in 1983. These fields are produced by existing power lines near the study plots. Plots were chosen to have minimal values for 60 Hz fields and to be matched as control and test plots, within the standard of one order of magnitude. Transverse electric fields were initially at or near the lower limits of measurability on all plots (Table 2). Low power testing of the antenna began in 1986 and continued through 1988 at increasing amperage. Values for transverse electric fields then increased on one test plot (1T2) in 1987, and the all test plots through 1990. Control plots remained unaffected. Apparently the fields produced from nearby power lines couple to the antenna and re-radiate as 60 Hz fields (Gauger, personal communication).

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Table 2. Mean values for 60 Hz transverse electric fields (V/m) on control and test plots paired by research activity. The values in parentheses are the sample N.

PLOT	1983-1985	1986	1987	1988	1989	1990	1991
Control							
1C1	0.001 (4)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)
1C3	0.001 (5)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)
1C4	0.001 (7)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)
1C6	0.001 (4)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)
Avg	0.001 (20)	0.001 (10)	0.001 (10)	0.001 (19)	0.001 (10)	0.001 (10)	0.001 (10)
Tree Swallow release site							
1D3	-	0.001 (1)	0.001 (1)	0.001 (1)	0.001 (1)	0.001 (1)	0.001 (1)
Test							
1T1	0.001 (11)	0.001 (14)	0.001 (14)	0.028 (18)	0.015 (8)	0.005 (18)	0.001 (11)
1T2	0.001 (3)	0.001 (4)	0.046 (5)	0.013 (5)	0.004 (5)	0.017 (5)	0.027 (5)
1T4	0.001 (4)	0.001 (5)	0.001 (10)	0.014 (10)	0.002 (10)	0.031 (10)	0.037 (10)
1T5	0.001 (5)	0.001 (6)	0.001 (9)	0.037 (9)	*	0.046 (9)	0.068 (7)
1T6	0.001 (4)	0.001 (1)	0.001 (7)	0.078 (7)	*	0.037 (7)	*
Avg	0.001 (27)	0.001 (30)	0.025 (45)	0.034 (49)	0.007 (23)	0.027 (49)	0.029 (34)
Tree Swallow release sites (Averaged)							
1D1 & 1D2	-	1.251 (2)	0.001 (2)	4.601 (2)	0.372 (2)	0.678 (2)	1.254 (2)

* = measurement precluded by antenna operation.

Averaged values for longitudinal electric and magnetic 60 Hz fields (Table 3 and Table 4) were higher on test compared to control plots in most years. Control test plot ratios varied from about 1 to over 27 fold for longitudinal fields, with the high value coming from 1988 for 1T6 vs 1C4 (Table 3). Longitudinal electric fields averaged highest on control plots in 1984 and on test plots in 1988. Magnetic fields remained relatively constant on controls but increased from 1986 through 1988 and then appear steady to 1990. On test plots, magnetic fields increased from 1986 to 1988 and then decreased in 1989, and 1990 (Table 4).

Among sites within the control plot, 1C1 and 1C3 (Michigamme North and South) were consistently higher for 60 Hz longitudinal electric fields (Table 3). Test plots 1T5 and 1T6 (Ford River North and South) were higher than other test sites in most years. Magnetic fields show larger values for site 1C6 but no patterns in other control plots. Plots 1T2-1T6 all increase in 1986, 1987, and 1988 (Table 4) but then decreased in 1989 and 1991. Site 1T1 shows a smaller increase and then a decrease in these years.

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Table 3. Mean values for 60 Hz longitudinal electric fields (mV/m) on test and control plots for years 1983 to 1991. The values in parentheses are the sample N.

PLOT	1983-1985	1986	1987	1988	1989	1990	1991
CONTROL							
1C1	0.093 (5)	0.100 (2)	0.114 (2)	0.338 (2)	0.137 (2)	0.056 (2)	0.082 (2)
1C3	0.158 (5)	0.080 (2)	0.148 (2)	0.117 (2)	0.178 (2)	0.110 (2)	0.102 (2)
1C4	0.039 (7)	0.065 (3)	0.047 (3)	0.048 (3)	0.024 (3)	0.022 (3)	0.036 (2)
1C6	0.079 (4)	0.068 (3)	0.089 (3)	0.041 (3)	0.079 (3)	0.066 (3)	0.062 (3)
Avg	0.092 (20)	0.076 (10)	0.100 (10)	0.136 (10)	0.105 (10)	0.064 (10)	0.069 (9)
Tree Swallow homing release site							
1D3	-	0.052 (1)	0.156 (1)	0.053 (1)	0.290 (1)	0.260 (1)	0.103 (1)
TEST							
1T1	0.116 (11)	0.070 (14)	0.070 (14)	0.252 (18)	0.080 (8)	0.068 (18)	0.060 (18)
1T2	0.196 (3)	0.074 (4)	0.059 (5)	0.075 (5)	0.047 (5)	0.051 (5)	0.057 (5)
1T4	0.174 (4)	0.086 (5)	0.076 (10)	0.110 (10)	0.046 (10)	0.167 (10)	0.100 (10)
1T5	0.253 (5)	0.079 (6)	0.078 (9)	0.159 (9)	*	0.181 (9)	0.136 (9)
1T6	0.569 (3)	0.230 (1)	0.297 (7)	1.324 (7)	*	0.443 (12)	0.272 (11)
Avg	0.262 (26)	0.080 (30)	0.108 (45)	0.384 (49)	0.058 (23)	0.187 (54)	0.124 (53)
Tree Swallow homing release sites							
1D1 & 1D2		5.035 (2)	1.280 (2)	0.715 (2)	1.695 (2)	1.275 (2)	1.540 (2)
(Averaged)							

* = measurement precluded by antenna operation.

Table 4. Mean values for 60 Hz magnetic fields (mG) on test and control plots for years 1983 to 1991. The values in parentheses are the sample N.

PLOT	1983-1985	1986	1987	1988	1989	1990	1991
CONTROL							
1C1	0.001 (4)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)
1C3	0.002 (5)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)
1C4	0.002 (7)	0.001 (3)	0.002 (2)	0.001 (3)	0.001 (3)	0.002 (3)	0.001 (3)
1C6	0.003 (4)	0.003 (3)	0.003 (3)	0.002 (3)	0.003 (3)	0.003 (3)	0.002 (3)
Avg	0.002 (20)	0.002 (10)	0.002 (9)	0.001 (10)	0.002 (10)	0.002 (10)	0.001 (10)
Tree Swallow homing release site							
1D3	-	0.003 (1)	0.002 (1)	0.002 (1)	0.013 (1)	0.009 (1)	0.009 (1)
TEST							
1T1	0.003 (11)	0.009 (14)	0.010 (14)	0.052 (18)	0.018 (8)	0.008 (18)	0.012 (18)
1T2	0.001 (3)	0.025 (4)	0.018 (5)	0.010 (5)	0.006 (5)	0.018 (5)	0.019 (5)
1T4	0.001 (4)	0.012 (5)	0.021 (10)	0.018 (10)	0.007 (10)	0.033 (10)	0.031 (10)
1T5	0.001 (5)	0.018 (6)	0.026 (9)	0.047 (9)	*	0.038 (9)	0.048 (9)
1T6	0.001 (3)	0.020 (1)	0.033 (7)	0.094 (7)	*	0.040 (12)	0.024 (12)
Avg	0.001 (26)	0.014 (30)	0.020 (45)	0.044 (49)	0.010 (23)	0.026 (54)	0.025 (54)
Tree Swallow release sites							
1D1 & 1D2	-	0.057 (2)	0.080 (20)	0.023 (2)	0.078 (2)	0.073 (2)	0.130 (2)
(Averaged)							

* = measurement precluded by antenna operation.

The control release location (1D3) and Panola Plains (1C4) control site

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for tree swallow homing shows small differences in field strength for electric and magnetic fields (Tables 2-4). However, much larger ratios appear on test release locations (1D1 and 1D2) and their corresponding test sites (1T2, 1T4) for transverse, longitudinal and magnetic fields (Tables 2-4).

Assessments of 60 Hz fields were also conducted in the laboratory where the actual measurements of maximal metabolic scope were taken. Fields at locations near equipment and in the outdoor animal holding facilities were

Table 5. 60 Hz air electric field intensities (V/m) measured at the laboratory where maximal aerobic metabolism was under study.

Site No., Meas. Pt.	1986	1987	1988	1989	1990	
					Before Shielding	After
1L1-1	/	--	--	--	--	--
1L1-2	0.94	0.96	--	--	--	--
1L1-3	0.79	0.034	/	/	/	0.58
1L1-4	0.042	0.047	0.062	/	/	/
1L1-5	-	-	-	/	/	/
1L1-6	-	-	-	/	/	/
1L1-7	-	-	-	8.1	8.5	1.34
1L1-8	-	-	-	0.88	0.76	0.037
1L1-9	-	-	-	60.0	18.1	3.90*
1L1-10	-	-	-	-	/	0.010

- = measurement point not established. -- = measurement point dropped.
/ = data not taken. * = 4.0 V/m with humidifier on.

measured by IITRI personnel. Magnetic field shielding was designed and provided by IITRI for animal metabolic chambers which were placed in a water bath during metabolic runs. The water bath's compressor, electric motor and pump were also shielded. The shielding and ground of lights, desks and large equipment significantly reduced the strength of magnetic and electric fields (Table 5 and Table 6).

76 Hz Fields. In 1986, 1987 and 1988, measurements were made on 76 Hz fields produced by the antenna during testing. Variation of 76 Hz fields was examined among control plots to see if they were homogeneous. Control plots were all uniform with respect to transverse electric (Table 7) and magnetic

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Table 6. 60 Hz magnetic flux densities (mG) measured at the laboratory where maximal aerobic metabolism was under study.

Site No., Meas. Pt.	1986	1987	1988	1989	1990
1L1-1	9.13	--	--	--	--
1L1-2	0.179	0.156	--	--	--
1L1-3	0.080	0.143	/	/	0.071
1L1-4	0.114	0.118	0.080	0.075	/
1L1-5	-	-	-	14.1 ^a	5.200 ^c
				21.0 ^b	0.620 ^d
1L1-6	-	-	-	3.2 ^a	2.400 ^c
				44.0 ^b	0.195 ^d
					0.081 ^e
1L1-7	-	-	-	0.65	1.69
1L1-8	-	-	-	1.46	0.88
1L1-9	-	-	-	48.0	0.86
1L1-10	-	-	-	-	0.75

- ^a measurement made in vertical orientation only in an open, unshielded can, submerged to its rim.
- ^b measurement made above the bath surface.
- ^c measurement made in closed, unshielded, fully submerged can.
- ^d measurement made in closed, shielded, fully submerged can.
- ^e measurement made in closed, shielded, fully submerged can with motor and pump shielding.
- measurement point not established.
- measurement point dropped.
- / data not taken.

fields (Table 8). For longitudinal electric fields sites 1C1 and 1C3 were significantly greater than 1C4 and 1C6.

Among test plots, 1T5 was greater than other sites for transverse electric fields and 1T6 was greater than other sites for longitudinal electric fields. No other patterns emerged. The control plots 1C1 and 1C3 are closer to the antenna system by several km, perhaps explaining their higher values. Test site longitudinal electric fields differ from each other because of varying distances to the antenna wire and because of variations in soil conductivity between and across sites.

Longitudinal electric and magnetic 76 Hz fields were significantly different for test and controls indicating that low amperage testing produced a "treatment" condition on test plots, compared to controls. This raises the

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question as to when the antenna was actually operational. We must know this in order to group data based on antenna operational status.

Table 7. Mean values for 76 Hz transverse electric fields (V/m) on test and control plots for years 1986 (4 amperes), 1987 (15 amperes), 1988 (75 amperes), 1989 - 1991 (150 amperes).

PLOT	TRANSVERSE FIELDS (V/m)					
	1986 (4 amps)	1987 (15 amps)	1988 (75 amps)	1989 (150 amps)	1990 (150 amps)	1991 (150 amps)
CONTROL						
1C1	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)
1C3	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)	0.001 (2)
1C4	0.001 (2)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)
1C6	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)	0.001 (3)
Avg	0.001 (9)	0.001 (10)	0.001 (10)	0.001 (10)	0.001 (10)	0.001 (10)
Tree Swallow homing release site						
103	0.001 (1)	0.001 (1)	0.001 (1)	0.001 (1)	0.001 (1)	0.001 (1)
TEST						
1T1	0.078 (14)	0.264 (14)	0.897 (18)	1.834 (18)	2.000 (18)	2.000 (18)
1T2	--	0.301 (5)	1.710 (5)	2.540 (5)	2.819 (5)	2.487 (5)
1T4	0.140 (5)	0.424 (10)	1.936 (10)	3.851 (10)	4.373 (10)	4.948 (10)
1T5	0.283 (5)	0.790 (9)	3.614 (9)	6.531 (9)	11.002 (9)	7.998 (9)
1T6	0.182 (1)	0.544 (7)	2.458 (7)	5.275 (7)	7.070 (7)	7.272 (7)
Avg	0.171 (25)	0.465 (45)	2.123 (49)	4.006 (49)	5.453 (49)	4.506 (49)
Tree Swallow release sites						
101&102 (Averaged)	0.001 (2)	0.001 (2)	0.001 (2)	0.009 (2)	0.012 (2)	0.014 (2)

-- = measurement point not established.

The release sites for tree swallow homing studies compared to their respective study plots show low ratios for control sites and higher ratios for test (Tables 7-9). Ratios generally increase from 1986 to 1991, although transverse fields show a drop in 1988 and increase again in 1989 and 1991.

Comments on Ambient Monitoring

We have elected to use weather station data from several nearby sites to monitor the effects of climatic conditions impinging on the plots. The plots are relatively close to each other and therefore experience the same major weather patterns. Differences probably exist due to variations in storm tracks, local topography and vegetative features. These differences will produce some degree of variability in response in our study animals, but in most cases we expect this to be small and random in direction. It is therefore our judgment that the greatest value of weather data will be for

Table 8. Mean values for 76 Hz longitudinal electric fields (mV/m) on test and control plots for years 1986 (4 amperes), 1987 (15 amperes), 1988 (75 amperes), 1989 - 1991 (150 amperes).

PLOT	LONGITUDINAL FIELDS (mV/m)					
	1986 (4 amps)	1987 (15 amps)	1988 (75 amps)	1989 (150 amps)	1990 (150 amps)	1991 (150 amps)
CONTROL						
1C1	0.021 (1)	0.085 (2)	0.430 (2)	1.505 (2)	1.185 (2)	1.215 (2)
1C3	0.022 (1)	0.068 (2)	0.335 (2)	0.960 (2)	0.895 (2)	0.945 (2)
1C4	0.001 (1)	0.003 (3)	0.013 (3)	0.046 (3)	0.044 (3)	0.046 (3)
1C6	0.001 (1)	0.005 (3)	0.020 (3)	0.079 (3)	0.074 (3)	0.062 (3)
Average	0.011 (4)	0.040 (10)	0.200 (10)	0.648 (10)	0.550 (10)	0.464 (10)
Tree Swallow homing release site						
1D3	0.008 (1)	0.053 (1)	0.210 (1)	0.850 (1)	0.890 (1)	0.630 (1)
TEST						
1T1	1.089 (14)	4.244 (14)	19.900 (18)	40.606 (18)	39.433 (18)	39.778 (18)
1T2	--	7.500 (5)	34.600 (5)	76.200 (5)	73.600 (5)	77.800 (5)
1T4	2.162 (5)	7.390 (10)	36.300 (10)	74.400 (10)	72.300 (10)	73.100 (10)
1T5	1.670 (5)	6.600 (9)	28.444 (9)	63.222 (9)	62.333 (9)	63.444 (9)
1T6	5.400 (1)	18.457 (7)	83.857 (7)	162.286 (7)	181.714 (7)	138.714 (7)
Average	2.580 (25)	8.838 (45)	40.620 (49)	83.343 (49)	85.876 (49)	68.939 (49)
Tree Swallow homing release sites						
1D1&1D2 (Averaged)	0.068 (2)	0.320 (2)	1.365 (2)	8.650 (2)	8.150 (2)	7.950 (2)

-- = measurement point not established.

examination of year to year effects, rather than within a year among plots.

There is one instance where we have become aware of an effect that is probably based on micro-climatic differences among the plots. The abundance of aerial insects that are preyed upon by tree swallows is greater on control plots (see section on tree swallow growth). However, test plots may be less affected by cold weather due to the adjacent forest than control plots. We instituted a program to sample aerial prey, in cooperation with Dr. D. Hussell in Ontario, Canada, in 1986. The program is given in greater detail below in the sections dealing with population statistics and growth of tree swallows. Last year we reported the first results in the section on tree swallow growth.

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Table 9. Mean values for 76 Hz magnetic fields (mG) on test and control plots for years 1986 (4 amperes), 1987 (15 amperes), 1988 (75 amperes), 1989 - 1991 (150 amperes).

PLOT	MAGNETIC FIELDS (mG)					
	1986 (4 amps)	1987 (15 amps)	1988 (75 amps)	1989 (150 amps)	1990 (150 amps)	1991 (150 amps)
CONTROL						
1C1	0.001 (1)	0.001 (2)	0.003 (2)	0.007 (2)	0.007 (2)	0.007 (2)
1C3	0.001 (1)	0.001 (2)	0.003 (2)	0.008 (2)	0.008 (2)	0.007 (2)
1C4	0.001 (1)	0.001 (3)	0.001 (3)	0.002 (3)	0.002 (3)	0.002 (3)
1C6	0.001 (1)	0.001 (3)	0.001 (3)	0.004 (3)	0.004 (3)	0.003 (3)
Avg	0.001 (4)	0.001 (10)	0.002 (10)	0.005 (10)	0.005 (10)	0.004 (10)
Tree Swallow homing release site						
1D3	0.001 (1)	0.001 (1)	0.002 (1)	0.008 (1)	0.008 (1)	0.004 (1)
TEST						
1T1	0.143 (14)	0.530 (14)	2.251 (18)	4.921 (18)	4.593 (18)	4.410 (18)
1T2	--	1.164 (5)	5.538 (5)	11.800 (5)	10.860 (5)	10.520 (5)
1T4	0.278 (5)	1.050 (10)	5.410 (10)	10.700 (10)	10.160 (10)	9.890 (10)
1T5	0.408 (5)	1.409 (9)	6.600 (9)	13.678 (9)	13.256 (9)	12.533 (9)
1T6	0.400 (1)	1.043 (7)	4.889 (7)	9.843 (7)	10.114 (7)	9.700 (7)
Average	0.307 (25)	1.039 (45)	4.938 (49)	10.188 (49)	9.797 (49)	8.400 (49)
Tree Swallow homing release sites						
1D1&1D2	0.001 (2)	0.002 (2)	0.007 (2)	0.090 (2)	0.105 (2)	0.121 (2)
(Averaged)						

-- = measurement point not established.

STUDY OF SMALL MAMMAL COMMUNITIES

I. Purpose

The small mammal community study has not been conducted since 1988. As we have stated in previous annual reports, differences between plots from year to year appeared to be site specific and variable. Animal population levels are inherently variable and such characteristics are not amenable to the examination of ELF effects within the levels of our stated statistical goals. The year to year variability would not allow us to adequately detect possible effects due to electromagnetic fields. Therefore, this study was a reasonable element to drop when budget constraints necessitated restructuring our research priorities.

PARENTAL AND NESTLING BEHAVIOR, AND FECUNDITY,
GROWTH AND MATURATION STUDIES - TREE SWALLOWS

I. Purpose

The purpose of these studies is to characterize several aspects of the reproductive process in tree swallows at test and control sites and assess possible effects of the ELF Communication System on these variables. Specifically, the following aspects of the reproductive process are compared between test and control sites and for each site from year to year: numbers of eggs per clutch, hatching success within clutches, fledging success, rates of growth and development of hatchlings, and nestling mortality rates. All of these work elements are described together in this one section because they are all conducted on the same populations of birds.

II. Methods

These studies were conducted in natural or artificial clearings where we have erected arrays of nest boxes. The boxes were made of cedar lumber and mounted on posts, 1.5m above the ground. Tree swallows readily elected to nest in the boxes, although higher occupancy rates were recorded on control plots (Figure 2; The data in this figure are based on five test plots containing a total of 165 nest boxes, and two control plots containing 239 nest boxes). We think this is due to the larger area of the control plots and relatively smaller edge of unsuitable vegetation. Adults at the nest tolerated considerable disturbance by investigators. The boxes could be opened to permit inspection and weighing of young. Sheets of high-density polyethylene wrapped around the posts prevented access by terrestrial predators.

When possible, adults were captured on the nest after incubation was completed and banded with U. S. Fish and Wildlife Service bands for identification. In addition, as many young as possible were banded before fledging.

Active nests were generally checked daily or every other day to determine the dates that eggs are laid, how many are laid, the dates the young hatch, and overall hatching success. During hatching, nests were checked twice

daily, to determine time of hatching with greater accuracy as well as the spread of hatching over time. Monitoring of the nests for nestling growth and mortality then continued until all young reached 15-16 days of age. Young tend to fledge unusually early if disturbed beyond day 16. Therefore, to minimize disturbance after day 16, nest checks to estimate fledging success were done every other day.

For studies of growth and development, nestlings were weighed every other day with a Pesola spring scale accurate to 0.1 g. The lengths of the tarsus, ulna, and wing (all from the right side of the body) were measured with dial calipers accurate to 0.1 mm. Since it was impossible for one observer to measure all nestlings we had at least two observers collecting growth data. However, we have noticed that different observers differ slightly in their techniques for measuring weights and body parts. Therefore we had all observers rotate among the plots so that every nestling was eventually measured by all observers. Regularly rotating the observers in this way has the effect of submerging the variance in measurement, due to observers, into the error in each nestling's growth curve. This measurement protocol unfortunately prevents us from being able to block observer effects in the statistical design. However, as we show below, when we use data from each individual bird's growth curve, even the significant effects of differences in observer technique do not prevent us from being able to detect small differences in patterns of growth.

For analysis of growth data, we used the procedure for fitting growth data to models of growth proposed by Ricklefs (1967, 1983) and used previously for tree swallows by Zach and Mayoh (1982). Briefly, the data for each nestling were subjected to curve fitting using an exponential or logistic

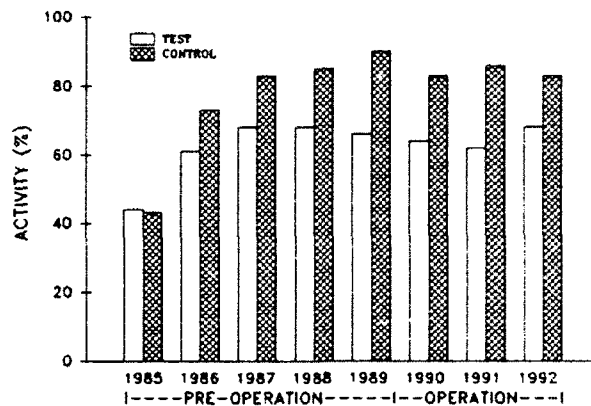


Figure 2. Egg laying activity (%) of Tree Swallows as a percentage of nestboxes occupied on test and control sites for 1985 through 1992.

model in a non-linear curve fitting routine in SYSTAT (Wilkinson 1988, 1990). The model of best fit, as judged by having the highest value of R^2 , was used in subsequent analyses to obtain the rate of growth, the intercept, and the inflection point. The model of best fit every year, including 1991, has been the logistic. We also tested values for maximum size attained for weight, tarsus and ulna (wing is still growing at fledging). We also computed a linear growth rate for ages 3 to 11 days (the period of linear growth) to compare to our other measures of growth. These measures have been shown by Zach (1988) to be less variable than fitted values, as our data also show.

In past years we have detected significant differences in growth rates of young tree swallows between test and control plots. Recent evidence suggests that food availability on a plot can have a significant effect on both clutch sizes and growth rates of tree swallows (Hussell and Quinney 1987, Quinney et al. 1986). In order to determine what degree of variation between test and control plots in growth rates is the result of food resource availability, we have undertaken steps to quantify the flying insect abundance at each site. We have worked with Dr. David Hussell of the Ontario Ministry of Natural Resources and have implemented a sampling scheme based on his earlier work (see Hussell and Quinney 1987, for detailed methodology). Briefly, we collected flying insects during the daylight hours in two suspended conical nets with alcohol traps at test and control plots used in the growth studies. These nets were stationed among the nest boxes and were constructed to face passively in the wind to continually sample insects which either fly or are blown into the nets. Previous studies have shown a reasonable relationship between the insects collected in this type of system and the insects delivered to young swallows in the nest by their parents (Quinney and Ankney 1985). Sampling began before the initiation of any egg laying and ended when all young from the plot had fledged. After insects were sorted into size classes we computed an index of the biomass of flying insects determined from daily catches on each plot.

Since we began the insect work, we have not found significant differences among plots for the variables of growth and maturation we measure. Therefore,

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we have not found any use for the insect measures in dealing with potential plot effects. There are effects due to weather, which we note as year effects. However, year effects are not of interest in assessing a possible antenna operation effect. Accordingly, we do not present any data on insect abundance in this report.

Nestling Transfer Experiment. In 1990 and 1991, we conducted a nestling transfer experiment to test for effects of short term exposure to ELF radiations. The rationale for the experiment was that one of the most powerful tests of ELF effects possible in our present research configurations would be to select a subset of individuals from the same nest and have them grow to independence in a nest on a corresponding test or control plot. Thus, individuals hatched on the test plot and transferred to the control plot would experience ELF radiations only as eggs, and nestlings transferred to the test plot from the control would only have experienced ELF radiations as nestlings. Controls were established for the effect of being raised by different parents and for transferring the young out of the nest. The procedure was to select nests at the same stage of development (they had to hatch on the same day) and assign them randomly to a control group, within plot exchange or across plot exchange. Next, nestlings were toe-marked with ink to indicate nestling 1 through 5 (all nests were standardized to five nestlings). Then a random set of three nestlings were chosen to be the transferred ones; the other two nestlings remained in their nest. In the control situation, the set of three selected nestlings were taken from the nest, but then immediately replaced, simulating the same procedure of moving nestlings among nests. In the within plot exchanges, the set of three nestlings were exchanged with another nest on the same plot. In this case, nestlings were either unexposed (control plot) but exchanged, or exposed (test plot) but exchanged for their entire nestling life. In the across plot exchanges, the set of three nestlings were exchanged with a set from the opposite plot. In this case, a set hatched on the control plot would be transferred to the test plot and the set hatched on the test plot was transferred to the control plot. These individuals were not exposed as eggs but were as nestlings (control to test plot) or were exposed as eggs

but not as nestlings (test to control plot). In both years, the setup of the experiment and coding of young was done by supervisory project personnel. Workers measuring growth of the experimental young had no knowledge of which young had been exchanged and therefore were "blind" to the experimental designations. Growth statistics were later matched to young by use of codes set up in the original design. To summarize, the treatment levels in the design were: 1) no exposure with sham exchange, 2) no exposure with exchange, 3) exposure as eggs but nestlings raised without exposure, 4) not exposed as eggs but raised as nestlings with exposure, 5) full exposure with no exchange, 6) fully exposed with exchange with other exposed nests. The effect of being reared by different parents in exchanged young was included in the error term. We are continuing to consult with statisticians to see if a suitable model can be designed to account for the parental effect.

A Note on Analysis of Variance Tests. We report results from analyses of variance models on growth variables taking into account antenna operation, years, plot and nests. The model used has two main fixed effect terms: OPERATION and PLOT. There are two random nested effects: YEAR(OPERATION) and NEST(PLOT), and the other terms of the model include interactions of the main effects and the nested effects. The appropriate error term for testing main effects is complicated by the presence of the nested effects. We have followed the procedures in Zar (1984, pages 470-476) for determining the appropriate error term. In general, the error term for the PLOT effect was the NEST(PLOT) mean squares, and for OPERATION, YEAR(OPERATION) mean square. Degrees of freedom were estimated using the formula provided by Zar (1984, page 473).

III. Results - 1992

The age of adult females breeding on the plots has been quantified as frequently as possible during all years of the study. First year females are distinct in that they exhibit a duller, distinctly brownish plumage as compared to the bright iridescent blue of older females (Hussell 1983). First year females also begin nesting slightly later and lay smaller clutches than

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older females (DeSteven 1979). The number of first year females settling on the plots has been variable from year to year, yet the majority are observed on the control plots. We are presently analyzing some of our data sets taking female age into account.

Fecundity. Summarized fecundity data for tree swallows in 1992 and previous years (1985-1991, Figure 3, Figure 4 and Figure 5) show that test and control plots appear to have nearly equal clutch sizes and hatching and fledging rates throughout the study. These data exclude any renesting attempts and analyses on fledging success exclude any nests manipulated for the reciprocal transplant growth studies in 1990 and 1991 (see tree swallow growth section).

Mean clutch sizes in 1992 were essentially the same at Pirlot Road test (5.3 eggs/nest) compared to clutches at Tachycineta Meadows control (5.2 eggs/nest, t-test, $t = 0.867$, $P = 0.388$). These values are within the range reported elsewhere for tree swallows (Chapman 1955, DeSteven 1979, Zach and Mayoh 1982, Hussell 1983b). In addition, there was no difference in the distribution of clutch sizes between test and control plot in 1992 ($\chi^2 = 1.066$, $P > 0.5$).

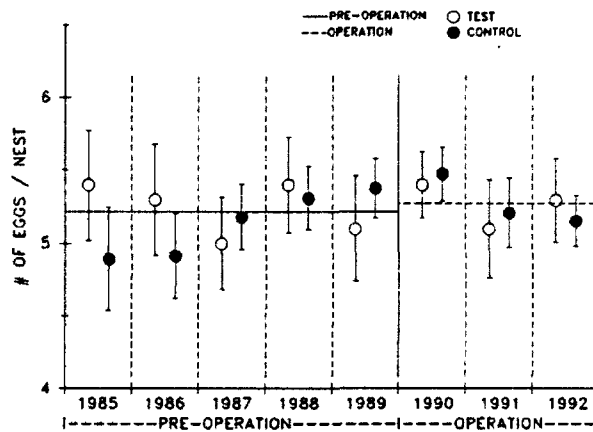


Figure 3. Clutch size (# of eggs/nest) of Tree Swallows observed on test and control sites for 1985 through 1992. Yearly plot means ($\pm 95\%$ confidence limits) and means for pre-operational (-), and operational (- -) antenna status are shown.

We analyzed data on clutch size from all years of the study (1985-1992) in a nested analysis of variance to assess the potential effects due to antenna OPERATION (preoperational 1985-1989, operational 1990-1992, as defined by IITRI), PLOT (test vs. control treatment plot), YEAR (nested within operation), and OPERATION*PLOT interaction. Due to the nested design, the error term used to compute the OPERATION effect F value was the YEAR (nested within operation) mean square. Results of this analysis (Table 10) show no

Table 10. Nested ANOVA for clutch size in tree swallows.

SOURCE	DF	TYPE III SS	MS	F	P
OPERATION	1	0.422	0.422	0.275	0.619
PLOT	1	0.232	0.232	0.343	0.559
YEAR(OPERATION)	6	9.223	1.537	2.267	0.036
OPERATION*PLOT	1	0.333	0.333	0.491	0.484
ERROR	631	427.815	0.678		

significant effects due to antenna operation or treatment plot, nor was any interaction detected. A significant effect of years within operational periods was detected ($F = 2.267$, $P = 0.036$). This appears to be due to yearly fluctuations in clutch size within antenna operation status, particularly once the antenna was activated (1990-1992).

The number of young hatched per nest in 1992 was not different between the Pirlot Road test plot (5.0 young/nest) and the Tachycineta Meadows control (4.8 young/nest $t = 0.868$, $P = 0.389$). These values for numbers of young hatched per nest are within the range reported elsewhere for tree swallows (Low 1933, Paynter 1954). We analyzed data on hatch rate from all years of the study (1985-1992, Figure 4) in a nested analysis of variance to assess the

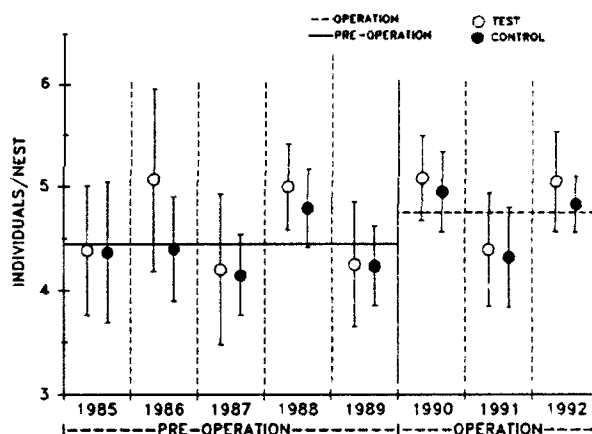


Figure 4. Hatch rate (individuals/nest) of nesting Tree Swallows observed on test and control sites from 1985-1992. Yearly plot means (\pm 95% confidence limits) and means for pre-operational and operational antenna status are shown.

potential effects due to antenna OPERATION (preoperational 1985-1989, operational 1990-1992), PLOT (test vs. control treatment plot), YEAR (nested within operation), and OPERATION*PLOT interaction. Due to the nested design, the error term used to compute the OPERATION effect F value was the YEAR (nested within operation) mean square. Results of this analysis (Table 11) show no significant effects due to antenna operation or treatment plot, nor

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was any interaction detected. As was the case with clutch size, a significant effect of year was detected ($F = 3.857$, $P = 0.001$). This year effect results from hatch rates being essentially equal between test and control plots in almost all years of the study, but overall hatch rates being higher in 1988, 1990, and 1992 and relatively lower in 1985, 1987, 1989, and 1991 (Figure 4).

Table 11. Nested ANOVA for hatch rate in tree swallows.

SOURCE	DF	TYPE III SS	MS	F	P
OPERATION	1	7.931	7.931	1.359	0.288
PLOT	1	2.568	2.568	1.697	0.193
YEAR(OPERATION)	6	35.021	5.837	3.857	0.001
OPERATION*PLOT	1	0.052	0.052	0.035	0.853
ERROR	437	661.347	1.513		

Another approach to the analysis of hatching success is to assess the likelihood to hatch, that is, the proportion of eggs which hatch, rather than using the number of eggs which hatch per nest as a continuous variate as was done in the nested analysis of variance. Using this approach, it was shown that the likelihood to hatch did not differ between test and control plots during 1992 (Table 12). On the test plot 96.4% of available eggs hatched, while 94.7 hatched on the control plot ($\chi^2 = 0.426$, $P = 0.514$). These values represent the highest percentages of eggs hatched during any year of the study.

Going beyond analysis at the yearly level we can first test for heterogeneity of the yearly 2×2 tables and also use multidimensional contingency tables to test hypotheses of mutual and partial independence (Zar 1984, Everitt 1977).

Considering the years 1985-1989 as preoperational antenna status and the years 1990-1992 as operational, we tested for heterogeneity of the yearly 2×2 tables to determine if these yearly samples of likelihood to hatch could be justifiably pooled within operational status (Zar 1984, pg. 67). This is done by first summing χ^2 values and degrees of freedom for each of the yearly samples within operational status. This value is termed the total χ^2 . The

Table 12. Likelihood of eggs to hatch for tree swallows from all years of the study, 1985-1992.

Data are from Pirlot Road test plot and Tachycineta Meadows control plot only and are compared using χ^2 tests.

Year	Plot	HATCHING SUCCESS			χ^2	P
		Hatch	Not Hatch	% Hatch		
1992	Test	106	4	96.4	0.426	0.514
	Control	198	11	94.7		
1991	Test	101	16	86.3	0.346	0.556
	Control	177	34	83.9		
1990	Test	122	7	94.6	2.064	0.151
	Control	203	22	90.2		
1989	Test	85	17	83.3	0.000	0.988
	Control	216	43	83.4		
1988	Test	90	8	91.8	0.283	0.595
	Control	206	23	90.0		
1987	Test	63	11	85.1	0.013	0.909
	Control	170	31	84.6		
1986	Test	71	6	92.2	2.972	0.085
	Control	132	25	84.1		
1985	Test	57	8	87.7	0.246	0.620
	Control	48	5	90.6		

data within operational status are then pooled over years and a χ^2 value is calculated for the resulting 2×2 table. This value is termed the χ^2 of totals. To calculate the heterogeneity χ^2 , the χ^2 of totals is subtracted from the total χ^2 and respective degrees of freedom are also subtracted. The resulting values of heterogeneity χ^2 can be compared to tabulated critical values with the resulting degrees of freedom. A rejection of the null hypothesis of homogeneity between the yearly samples within operational status indicates that the yearly samples are heterogeneous and, thus cannot be pooled.

Yearly likelihood to hatch values for preoperational antenna status ($\chi^2 = 2.429$, $df = 4$, $P > 0.5$) and operational antenna status ($\chi^2 = 0.717$, $df = 2$, P

> 0.5) were both found to be homogeneous. With yearly data pooled within operational status the hypothesis of mutual independence (Zar 1984, Everitt 1977) was then tested and rejected ($\chi^2 = 16.51$, $df = 4$, $P = 0.002$). From this we can conclude that some combination of treatment plot or antenna operation status is causing the lack of independence observed in likelihood to hatch values. The rejection of the hypothesis of mutual independence leads to three tests of partial independence:

- 1) likelihood to hatch is independent of treatment plot and operational status
- 2) treatment plot is independent of operational status and likelihood to hatch
- 3) operational status is independent of likelihood to hatch and treatment plot.

Hypotheses 1 and 3 were rejected (both $P < 0.01$), but hypothesis 2 was not rejected ($\chi^2 = 7.19$, $df = 3$, $P = 0.066$) which leads to the conclusion that treatment plot is independent of operational status and likelihood to hatch. Because this second hypothesis was not rejected it is desirable to pool data over treatment plot and test the hypothesis that antenna operation is independent of likelihood to hatch. This hypothesis was subsequently rejected ($\chi^2 = 9.262$, $df = 1$, $P < 0.005$) which leads to the conclusion that likelihood to hatch is not independent of antenna operation. This lack of independence is not due to the effects of antenna operation per se, but rather to major differences in years (weather) during antenna operation. Overall percentages of eggs which hatched increased following the start of antenna operation (86.5% preoperational, 90.6% operational), but this increase occurred on both test and control plots.

Due to prolonged inclement weather during mid-June and overnight freezing temperatures on June 19, 20 and 21, we experienced a very high rate of mortality of nestling tree swallows. The unusual freezing temperatures came at a time when hatching of young was complete for a majority of nests, but prior to the time when nestlings could effectively thermoregulate on their own. Several plots experienced greater than 95% mortality of young.

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Inclement weather caused similar high mortality in 1986 and 1989 (see Figure 5). The events in 1992 proved to be of longer duration with more extreme temperatures.

The numbers of young fledged per nest in 1992 were very low at both the Pirlot Road test plot (0.14 young/nest) and Tachycineta Meadows control (0.38 young/nest), and these values were not significantly different ($t = -0.841$, $P = 0.404$). These values represent the lowest numbers of young fledged per nest for any year of the study (Figure 5).

Table 13. Nested ANOVA for fledge rate in tree swallows.

SOURCE	DF	TYPE III SS	MS	F	P
OPERATION	1	7.335	7.335	0.070	0.800
PLOT	1	0.911	0.911	0.284	0.595
YEAR(OPERATION)	6	627.838	104.640	32.560	0.000
OPERATION*PLOT	1	2.433	2.433	0.757	0.385
ERROR	390	1253.346	3.214		

Data on mean fledging success from all years of the study (1985-1992, Figure 5) were analyzed in a nested analysis of variance to assess the potential effects due to antenna OPERATION (preoperational 1985-1989, operational 1990-1992), PLOT (test vs. control treatment plot), YEAR (nested within operation), and OPERATION*PLOT interaction. Due to the nested design, the error term used to compute the OPERATION effect F value was the YEAR (nested within operation) mean square. Results of this analysis (Table 13) show no significant effects due to antenna operation or treatment plot, nor was any interaction detected. As was the

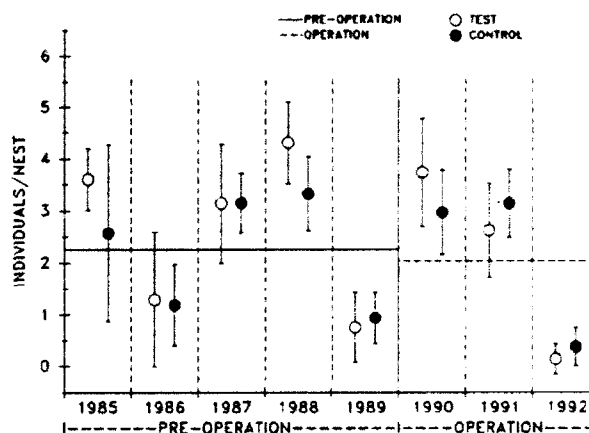


Figure 5. Fledge rate (individuals/nest) of nesting Tree Swallows observed on test and control sites for 1985-1992. Yearly plot means (\pm 95% confidence limits) and means for pre-operational and operational antenna status are shown.

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case with clutch size and hatching rate, a significant effect of year was detected ($F = 32.56$, $P < 0.001$) which is clearly a result of low fledging rates during the poor weather years of 1986, 1989 and 1992 (Figure 5).

The proportion of young fledged from those eggs which hatched during 1992, although very low, also did not differ between test (2.8%) and control

Table 14. Likelihood of young to fledge for tree swallows from all years of the study, 1985-1992.

Year	Plot	FLEDGING SUCCESS			χ^2	P
		Fledge	Not Fledge	% Fledge		
1992	Test	3	103	2.8	2.953	0.086
	Control	15	178	7.8		
1991	Test	55	36	60.4	6.012	0.014
	Control	116	38	75.3		
1990	Test	56	17	76.7	6.236	0.013
	Control	89	60	59.7		
1989	Test	15	70	17.6	0.784	0.376
	Control	47	164	22.3		
1988	Test	69	12	85.2	7.832	0.005
	Control	123	56	68.7		
1987	Test	44	17	72.1	0.927	0.336
	Control	126	35	78.3		
1986	Test	18	53	25.4	0.071	0.790
	Control	32	86	27.1		
1985	Test	36	7	83.7	6.012	0.014
	Control	18	13	58.1		

(7.8%) plots ($\chi^2 = 2.953$, $P = 0.086$, Table 14).

Going beyond analysis at the yearly level, as was done for hatching success, we can first test for heterogeneity of the yearly 2 X 2 tables and also use multidimensional contingency tables to test hypotheses of mutual and partial independence (Zar 1984, Everitt 1977). Yearly χ^2 values (Table 14) show significant lack of independence during 1985, 1988, 1990 and 1991. In

three of these four years, likelihood to fledge on test plots was greater than the controls.

Taking the same approach as used for analysis of hatching data, we tested for heterogeneity of the yearly 2 X 2 tables within antenna operational status. For likelihood to fledge during preoperational antenna status, yearly samples proved to be heterogeneous ($\chi^2 = 14.199$, $df = 4$, $P < 0.01$) and thus cannot be pooled. The same result is obtained during antenna operational years ($\chi^2 = 14.787$, $df = 2$, $P < 0.001$). Since yearly samples proved to be heterogeneous and could not be pooled, we tested hypotheses of mutual independence within antenna operation. For the preoperational years of 1985-1989 we rejected the hypothesis of mutual independence ($\chi^2 = 335.36$, $df = 13$, $P < 0.001$). Three tests of partial independence were also rejected (all $P < 0.001$). The conclusion here is that likelihood to fledge is influenced by some combination of year and treatment plot during preoperational antenna status. Two of the five years of preoperational status (1985 and 1988) show significant differences between test and control plots in proportions of young fledging and there is tremendous disparity between years due to the episodes of inclement weather in 1986 and 1989 (Table 14).

For the operational years of 1990-1992, the hypothesis of mutual independence was rejected ($\chi^2 = 292.38$, $df = 7$, $P < 0.001$), as well as all three tests of partial independence (all $P < 0.007$). Once again, we conclude that likelihood to fledge is dependent upon some combination of treatment plot and year during the years when the antenna was operational. There are significant differences between test and control plot during 1990 and 1991, yet these yearly differences are in opposite direction to one another. In addition overall likelihood to fledge in 1992 was very low on both test and control plots when compared to 1990 and 1991.

These contingency table results show that during both preoperational and operational antenna status, likelihood to fledge was highly variable and was influenced by differing combinations of treatment plot and year.

In conclusion, there were no significant differences between test and control plots during 1992 for clutch size, hatch rate, or fledge rate. The

nested analysis of variance on data from all years of the study likewise shows no effects due to antenna operation or treatment plot for these three variables. All three variables showed a significant effect of year. Data on likelihood to hatch shows a significant effect due to antenna operation following the pooling of years, but since no differences were exhibited between test and control plots, we interpret the difference as due to weather events. Proportion of eggs hatching increased following the start up of the antenna system, but the increase was shown on both test and control plots. Data on likelihood to fledge are much more difficult to interpret since it appears as though the counts of fledged versus not fledged are highly dependent upon both treatment plot as well as year within operational status.

Mortality. In addition to the intensive study of fecundity variables at the Pirlot Road test plot and Tachycineta Meadows control plot, we monitored all active nests at two additional test plots (North Turner and Cleveland Homestead) and one additional control plot (Panola Plains). Thus, three test plots pooled and two control plots pooled provide the basis for an analysis of overall nesting success based on the Mayfield method (Mayfield 1961, 1975). This method takes into account the exposure of each nest to potential mortality factors over the number of days the nest is under observation. The unit of exposure used for comparisons between groups is the nest-day. For example, one nest under observation for ten days represents ten nest-days, or five nests under observation for 15 days would sum to 75 nest-days of exposure. Therefore the total exposure of a group of nests (say, at a treatment plot) would be the summation of nest-days for each nest in the group. Other units of exposure which provide a finer-tuned approach are the egg-day and the nestling-day. For example, a nest with five eggs observed for eight days represents 40 egg-days of exposure as well as eight nest-days. Nestling-days are dealt with in the same manner. Most of the nests included in this analysis were observed from the day of the first egg laid to the completion of the nesting attempt. Any manipulated nests (for example from the 1990 and 1991 reciprocal transplant growth study) were excluded from any analysis of nestling mortality or nestling phase nest mortality.

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We use two methods to compare groups of nests. The traditional method as outlined by Mayfield (1975) employs contingency table analysis using counts from each group of nests of nest-days with mortality and nest-days without mortality. The method presented by Hensler and Nichols (1981) employs maximum likelihood estimates and variances of survival probabilities to compare groups. Here we present results from 1992 data using both methods (Table 15).

Table 15. Exposure data and frequency of NEST mortality throughout the entire nesting cycle during 1992 calculated using the Mayfield method (Mayfield 1961,1975).

OVERALL NEST MORTALITY						
Plot	days without nest failures	days with nest failures	χ^2	P	Z	P
Test	2253	90	13.523	0.000	3.317	0.000
Control	6079	148				
INCUBATION PHASE NEST MORTALITY						
Plot	days without nest failures	days with nest failures	χ^2	P	Z	P
Test	1574	25	0.646	0.422	0.773	0.221
Control	3763	49				
NESTLING PHASE NEST MORTALITY						
Plot	days without nest failures	days with nest failures	χ^2	P	Z	P
Test	679	65	25.608	0.000	4.220	0.000
Control	2343	99				

The analysis of overall nest mortality compares test and control values for the number of nest-days without mortality and the number of days where nest failure occurred throughout the entire nesting attempt. A significant

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lack of independence was detected between test and control plots in 1992 ($\chi^2 = 13.523$, $P < 0.001$); nest mortality being higher on the test plots (Table 15). The same result is obtained using the Hensler and Nichols method. Significant differences in survival probabilities were found ($Z = 3.32$, $P < 0.001$).

It is of interest to break down the overall nest mortality data into two periods. The first represents the time span of egg laying and incubation (hereafter termed incubation phase) and the second the period of time when eggs have hatched and young are being fed by the adults (hereafter termed nestling phase). These two time periods are characterized by different levels of parental care by both parents. Incubation of eggs, followed by brooding of the young for several days following hatching are female behaviors, yet the male and female later share approximately equal roles in bringing food to the young.

Incubation phase nest mortality in 1992 showed no differences between treatment plot using either method of analysis ($\chi^2 = 0.646$, $P = 0.422$; $Z = 0.773$, $P = 0.221$), whereas nestling phase nest mortality was shown to be significantly different between plots for both methods ($\chi^2 = 25.608$, $P < 0.001$; $Z = 4.22$, $P < 0.001$).

Using the egg-day as the unit of exposure and computing egg mortality we found no significant differences in mortality of individual eggs between test and control plots during 1992 using either method of analysis ($\chi^2 = 2.836$, $P = 0.092$; $Z = 1.632$, $P = 0.0516$). However, using the nestling-day as the unit of exposure and computing nestling mortality we found significantly higher mortality rates on the test plots when compared to the control plots during 1992 using either method ($\chi^2 = 70.032$, $P < 0.001$; $Z = 7.324$, $P < 0.001$) (Table 15).

In conclusion, during 1992 overall nest failure and nestling mortality rates were higher on the test plots. Although equal rates of nest failure and egg mortality were exhibited during the incubation phase, mortality was more severe on the test plots. It is possible the 1992 severe weather event was of different intensity or had different manifestations at sites for test and

Table 16. Exposure data and frequency of mortality of EGGS and NESTLINGS during 1992 calculated using the Mayfield method (Mayfield 1961,1975).

EGG MORTALITY						
Plot	days without mortalities	days with mortalities	χ^2	P	Z	P
Test	6845	162	2.836	0.092	1.632	0.052
Control	16173	325				

NESTLING MORTALITY						
Plot	days without mortalities	days with mortalities	χ^2	P	Z	P
Test	2767	298	70.032	0.000	7.324	0.000
Control	9201	531				

control plots. We plan on analyzing ambient temperature data from two test plots and one control plot to test this hypothesis. In addition, a multidimensional contingency table analysis which will include all years of the study is planned.

Landmark growth events. [Note: no data are presented for 1992 because of the nearly complete mortality of young due to bad weather.] The mean number of days to eye opening in 1991 (Figure 6 and Figure 6) was longer at the Pirlot Road test plot (5.1 days) than at Tachycineta Meadows control (4.7 days), however these differences were not significant in analysis of variance (Table 16, $P > 0.25$).

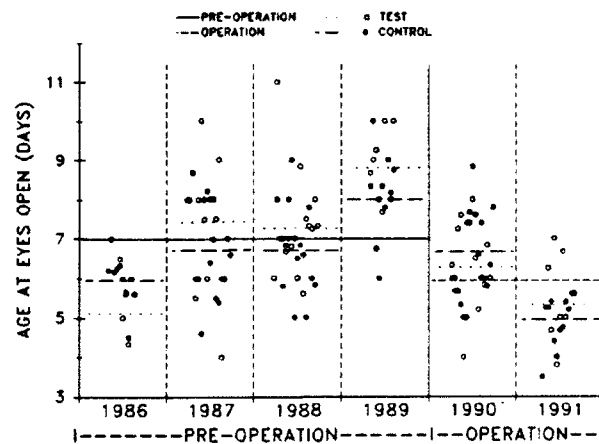


Figure 6. Age at eye opening (days) of nestling Tree Swallows observed on test and control sites for 1986 through 1991. Each dot is a nest mean.

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Neither was there a difference detected for OPERATION or PLOT*OPERATION (Table 16, $P = 0.2365$ and $P = 0.39$, respectively). We did detect a significant effect of YEAR(OPERATION) which we interpret as a weather effect, NEST(PLOT) due to variation of eye opening among nests, and PLOT*YEAR(OPERATION) which is a weather and plot interaction (Table 16, all $P < 0.01$). These factors are unrelated to ELF

radiation and appear to impact test and control plots equally. However, we are interpreting the of PLOT and YEAR interaction as due to weather effects on test plots caused by the closeness of the surrounding forest.

The scoring of eyes closed or open in the field is somewhat subjective and may be biased depending upon observer, lighting conditions and other factors. In addition, we only observe the young on an every-other-day basis. The resulting increase in the variance further reduces our ability to detect subtle differences in age of eye opening. Still, the differences in when nestlings achieve open eyes varies enough among nests and years that we consistently register significant differences. Differences among plots or operational periods are too small to be detected at the level of variation we currently have in these data.

Mean number of days to feather eruption in 1991 (Table 18) was similar to other years and very similar among plots (Table 20, $P > 0.386$). No significant effects of plot have been noted for any other years either (Table 17). As with age at eye opening, there is a significant effect of weather and of nest on the age at feather eruption (Table 17, YEAR(OPERATION), NEST(PLOT), $P = 0.0001$). In addition, a significant PLOT*OPERATION interaction occurred (Table 17, $P = 0.0012$). We feel this result is due to bad weather years which occurred in 1986 and 1989 during the pre-operational period and was more

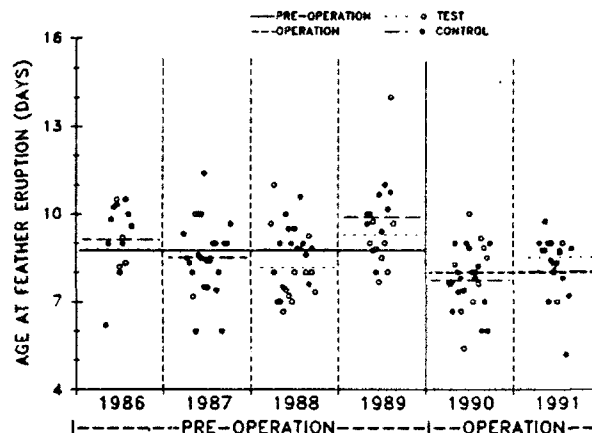


Figure 7. Age at feather eruption (days) of nestling Tree Swallows observed on test and control sites for 1986 through 1991. Each dot is a nest mean.

Table 17. Nested ANOVA for age of eye opening in tree swallows. Tested are the effects of PLOT (test or control), and nests (NEST) within a plot (PLOT) for 1986 through 1991.

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	145.7125	145.7125	1.94	0.2365
PLOT	1	2.1227	2.1227	0.18	0.25
YEAR(OPERATION)	4	301.0641	75.2660	46.54	0.0001
NEST(PLOT)	30	125.6928	4.1898	2.59	0.0001
OPERATION*PLOT	1	1.1675	1.1675	0.72	0.3959
PLOT*YEAR(OPERATION)	4	37.6160	9.4040	5.82	0.0001
ERROR	560	905.6020	1.6171		

severe on control plots due to their more exposed setting. No significant bad weather has occurred during the operational period (except 1992).

Comparing feather eruption with eye opening, the eruption of primary feathers is about as variable as eye opening (Table 18). It is much less subjective in the field when the actual scoring takes place. It is clear that variation in feather eruption is strongly influenced by weather and the nest (or parent) environment, but not ELF exposure.

Table 18. Nested ANOVA for primary feather eruption in tree swallows.

SOURCE	DF	SS	MS	F	P > F
PLOT	1	0.7378	0.7378	0.07	0.25
OPERATION	1	71.3894	71.3894	4.46	0.1023
YEAR(OPER)	4	64.0205	16.0051	12.94	0.0001
NEST(PLOT)	30	235.0503	7.8350	6.34	0.0001
PLOT*OPER	1	13.1221	13.1221	10.61	0.0012
ERROR	560	692.4505	1.2365		

Statistical sufficiency - fecundity and maturation. We have examined the statistical power of test and minimum detectable difference for the measures

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of fecundity and maturation discussed above. We are currently able to detect changes of less than 10% of the mean for the variables measured, but the power of these tests are very low. If we set the power of the test at 70% certainty, we are still able to detect differences of less than 10% for clutch size and hatch success, but only about 18% for fledging success (Table 19).

Table 19. Detectable differences and power for tree swallow fecundity variables.

Variable	N	Actual Detectable Difference (%)	Actual Power	Detectable Difference at 70% Power (%)
Clutch size eggs(%)	227	0.069(1.3)	<0.30	0.20(3.7)
Hatch success eggs(%)	155	0.029(0.6)	<0.30	0.35(6.9)
Fledging success young(%)	135	0.303(9.4)	<0.30	0.57(17.6)

Minimum detectable differences are larger for eye opening and feather eruption, but still are all below 17%, with power again less than 30%. With the power set to 70%, minimum detectable differences increase to greater than 25% and less than 45%, depending on the year (Table 20). We will therefore be less confident of rejecting the hypothesis of no difference in plots for these variables.

Adult Return Rates. In 1992, 252 adults were captured; 103 (41%) were new individuals and 149 (59%) were returning birds banded by us during previous seasons. The number of adults captured was considerably less than in previous years due to the high rate of abandonment caused by a cold and wet nesting season. The proportion of returning adults in 1992 was greater than previous years; 45.6% in 1992, 41.3% in 1990, 43.5% in 1989, 33.8% in 1988, 12.3% in 1987, 29.7% in 1986 and 16.6% in 1985. As many young as possible are banded before fledging; in 1992, only 210 young were banded in the nest compared to 828 young banded in 1991. In 1992, as in 1989 and 1986, nest

Table 20. Detectable differences and power for tree swallow landmark events; eye opening and feather eruption.

Variable	Year	N	Actual Detectable Difference(%)	Actual Power	Detectable Difference at 70% Power (%)
Eye opening days(%)	1991	9	0.83(17.0)	<0.30	2.20(45.0)
	1990	13	0.52(8.4)	<0.30	2.20(35.4)
	1989	8	0.83(10.1)	<0.30	2.08(25.5)
	1988	14	0.73(10.4)	<0.30	2.55(36.4)
	1987	12	0.43(6.2)	<0.30	3.15(45.1)
	1986	6	0.90(15.7)	<0.30	1.84(32.1)
Feather eruption days(%)	1991	9	0.465(5.8)	<0.30	2.70(34.0)
	1990	13	0.763(10.1)	<0.30	2.25(29.6)
	1989	8	0.885(9.4)	<0.30	2.85(30.4)
	1988	14	1.168(13.7)	<0.30	2.35(27.6)
	1987	12	1.009(11.9)	<0.30	2.65(31.2)
	1986	6	1.439(16.0)	<0.30	4.05(45.0)

abandonment by the adults and the high mortality of young caused by inclement weather reduced the number of birds available for banding. The low number of returning birds in 1987 during nesting may be a reflection of the 1986 cold weather. We have now accumulated enough data to begin assessing the return rates of adults to control and test plots. These results will be presented in future annual reports.

Growth. Curve fitting to growth data for individual birds during 1991 for body mass, tarsus and ulna growth was accomplished using the logistic model while wing growth was fit by the exponential model. These models produce the highest R^2 values, on average, compared to other growth models (see Ricklefs 1983, and Zach and Mayoh 1982, for discussion of various models). Linear regression of growth data for weight, tarsus and ulna between the ages of 3 and 11 days were dropped in 1992 since maximum values gave less variable results. The maximum values attained in growth for these variables and the age they were attained were also used to assess growth of nestlings on

control and test plots. As noted earlier, maximum values have been found by other researchers to be less variable than curve fitted ones (Zach, 1988), as we have also found. We include these more sensitive measures along with the curve fit values.

The logistic model was fitted to the data using a NONLIN procedure (Ricklefs 1983, Wilkinson 1988). The procedure estimates values for the growth rate constant and the inflection point for body mass, tarsus and ulna growth. The NONLIN procedure was also used to fit wing growth data to an exponential model of growth. A growth constant was estimated, but no inflection point occurred since the wing was still growing at the time of fledging. The growth and inflection point variables for each nestling were included in the data set if there was a significant correlation between the variable and age. The data were then analyzed using nested analysis of variance (NANOVA), with main effects of operational period and plot and their interaction and with the effect of years nested within period of operation and the effect of nests included within plots. Thus, the model may be written as:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + \alpha\beta_{ij} + \Gamma_{ik} + \Delta_{jl} + \Theta_{ij} + \Phi_{ijk} + e_{ijkl}$$

where Y_{ijkl} is the l th observation in the k th and j th subgroups of the i th group, μ is the parametric mean of the population, α_i is the fixed effect of the i th group (plots), β_j is the fixed effect of the j th group (operation period), and the other terms represent interaction of the main effects ($\alpha\beta_{ij}$), the nested effects of nest within plot and years within operation (Γ_{ik} , Δ_{jl}) and interaction of the nested effect of plot and years within operation (Θ_{ij}). e_{ijkl} is the error term. A nested model was used to account for the known effect of parents on the growth of their nestlings. Ricklefs and Peters (1981) studying the European starling (*Sturnus vulgaris*)

in Pennsylvania found the most significant contribution of variance to total variance in growth was due to the parents rather than variation in individual nestling growth or inherited growth traits. Our data on tree swallows shows similar partitioning of the variance in growth. We also include years as nested with operation since the years 1985-89 are entirely within the pre-operation period and the years 1990-92 are in the operation period. The appropriate mean square ratio for computing the value of F for a treatment (PLOT) effect is the mean square due to plot divided by the mean square due to nests within plot (Zar, 1984). This reduces the effective sample N to the number of nests within years and operation rather than the number of nestlings, and has some important impacts on the power of the test. This will be discussed in detail below after summarizing the findings for the years 1985 through 1991. Data are not available for 1992 since nearly all nestlings died in the cold weather episode.

In general, growth constants and inflection points from the curve fitted data and maximum values and ages at the maximum values were most strongly affected by nests within plots and least by plot. These results will be examined in turn below.

For body mass, growth constants, inflection points, maximum weight attained and age at maximum weight showed no significant plot or operational period effects (Table 21). Nest and year factors are highly significant for all measures. For nests, we find this is due to significant variation in the outcome of growth of body mass due to parental care (or inheritance?). The year effect is due to weather events that have a major impact on growth of mass when years are cold. We also detected an interaction of plot and operation for inflection point, but not the other weight variables. We are examining the means for these effects to see if we can interpret the meaning

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Table 21. Nested ANOVA for measures of body mass during growth in nestling tree swallows.

Nested ANOVA for weight growth constant for nestling tree swallows.

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	0.0022	0.0022	0.09	0.25
PLOT	1	0.0354	0.0354	0.75	0.4262
YEAR(OPER)	5	0.2362	0.0472	13.18	0.0001
NEST(PLOT)	42	0.7125	0.0170	4.73	0.0001
OPER*PLOT	1	0.0002	0.0002	0.06	0.8084
ERROR	660	2.3665	0.0036		

Nested ANOVA for the inflection point of fitted growth constants

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	10.2227	10.2227	0.63	0.4640
PLOT	1	0.0531	0.0531	0.01	0.25
YEAR(OPER)	5	81.4036	16.2807	23.96	0.0001
NEST(PLOT)	42	113.9758	2.7137	3.99	0.0001
PLOT*OPER	1	5.6665	5.6665	8.34	0.0040
ERROR	660	448.4399	0.6795		

Nested ANOVA for the maximum weight attained

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	3.8075	3.8075	0.13	0.7360
PLOT	1	15.9818	15.9818	0.59	0.25
OPER*PLOT	1	0.1076	0.1076	0.03	0.8701
YEAR(OPER)	5	149.8118	29.9624	7.45	0.0001
NEST(PLOT)	41	521.5729	12.7213	3.16	0.0001
ERROR	692	2781.8991	4.0201		

Nested ANOVA for the age at maximum weight

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	5.1431	5.1431	0.14	0.7207
PLOT	1	0.2501	0.2501	0.019	0.50
YEAR(OPER)	5	179.6051	35.9210	18.45	0.0001
NEST(PLOT)	41	341.5474	8.3304	4.28	0.0001
PLOT*OPER	1	2.5299	2.5299	1.30	0.2547
ERROR	693	1349.1232	1.9468		

of the interaction. Examples of body mass growth constants (Figure 8) and maximum weight (Figure 12) are shown as examples of patterns in the means.

For growth of the tarsus (Table 22), we also find no effect due to plot or operation period. Highly significant effects due to nest and year are

again present. We interpret these effects the same as for weight growth. Interaction of plot and operation show mixed significance depending on the variable. We are presently examining the means for these variables to interpret the meaning of the interaction. Examples of tarsus growth constants (Figure 9) and maximum tarsus length (Figure 13)

are shown as examples of patterns in the means.

An examination of ulna growth indicates that there were no plot or operational period effects (Table 23). We again find consistent, highly significant effects due to nest and year. The interaction of plot and operation is significant for some ulna variables and not others. The interaction of plot and years is highly significant for all but the growth constant. We are presently examining the means for these variables to interpret the meaning of these interactions. Examples of ulna growth constants (Figure 10) and maximum ulna length (Figure 14) are shown to illustrate patterns in the means.

Growth of the wing was examined by fitting data to an exponential model to produce a growth constant. The wing does not have a linear phase during growth in the nest and growth is still underway when nestlings fledge. Accordingly, we have only measures of the fitted growth constant to examine for possible ELF effects (Table 24). No effects of plot or operation were detected, but as for the other growth measures, significant effects for nest and year were detected as well as interactions of plot with operation plot

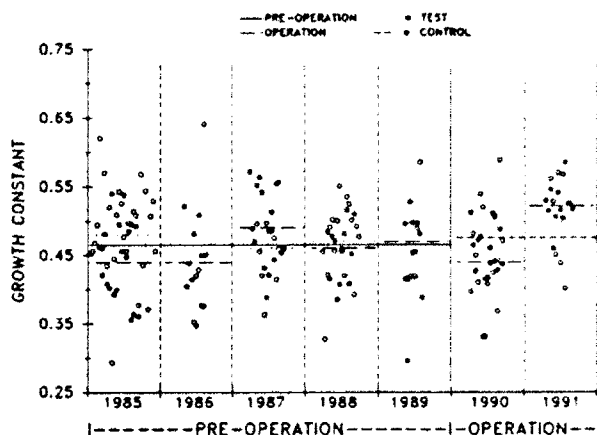


Figure 8. Weight growth constants (grams/day) of nestling Tree Swallows observed on test and control sites for 1985 through 1991.

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Table 22. Nested ANOVA of measures of tarsus growth in nestling tree swallows.

Nested ANOVA for tarsus growth constant

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	0.0624	0.0624	0.27	0.6259
PLOT	1	0.0089	0.0089	0.11	0.25
YEAR(OPER)	5	1.1575	0.2315	33.90	0.0001
NEST(PLOT)	43	1.5901	0.0370	5.42	0.0001
PLOT*OPER	1	0.0607	0.0607	8.89	0.0030
ERROR	699	4.7733	0.0068		

Nested ANOVA for the inflection point of tarsus growth

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	24.8794	24.8794	1.57	0.2658
PLOT	1	0.2484	0.2484	0.05	0.25
YEAR(OPER)	5	79.3138	15.8628	42.34	0.0001
NEST(PLOT)	43	84.3696	1.9621	5.24	0.0001
PLOT*OPER	1	9.2802	9.2802	24.77	0.0001
ERROR	699	261.8972	0.3747		

Nested ANOVA for the maximum length of tarsus

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	13.2034	13.2034	0.75	0.4257
PLOT	1	0.0061	0.0061	0.003	0.50
OPER*PLOT	1	0.5469	0.5469	1.46	0.2276
YEAR(OPER)	5	87.8889	17.5778	46.88	0.0001
NEST(PLOT)	43	28.3890	0.6602	1.76	0.0023
ERROR	712	266.9928	0.3750		

Nested ANOVA for the age at maximum length of tarsus

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	20.7155	20.7155	0.73	0.4326
PLOT	1	0.4303	0.4303	0.005	0.50
YEAR(OPER)	5	142.3325	28.4665	5.53	0.0001
NEST(PLOT)	43	568.8530	13.2291	2.57	0.0001
PLOT*OPER	1	64.3782	64.3782	12.50	0.0004
ERROR	712	3666.7050	5.1499		

with years (Figure 11).

Summary of growth measures. The data on growth consistently show there is no detectable effect due to exposure to ELF electromagnetic fields on plots

Table 23. Nested ANOVA for measures of the growth of the ulna in nestling tree swallows.

Nested ANOVA for ulna growth constant in tree swallow nestlings

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	0.0002	0.0002	0.00	0.9499
PLOT	1	0.0049	0.0049	0.52	0.25
YEAR(OPER)	5	0.2078	0.0416	18.77	0.0001
NEST(PLOT)	34	0.2853	0.0084	3.79	0.0001
PLOT*OPER	1	0.0089	0.0089	4.01	0.0458
ERROR	641	1.4192	0.0022		

Nested ANOVA for the inflection point of ulna growth

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	26.7691	26.7691	1.17	0.3279
PLOT	1	0.2992	0.2992	0.04	0.25
YEAR(OPER)	5	113.9435	22.7887	35.64	0.0001
NEST(PLOT)	34	82.0714	2.4139	3.78	0.0001
PLOT*OPER	1	7.7648	7.7648	12.14	0.0005
ERROR	641	409.8627	0.6394		

Nested ANOVA for the maximum length of ulna

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	7.1847	7.1847	0.20	0.6762
PLOT	1	22.4976	22.4976	0.93	0.25
OPER*PLOT	1	0.0225	0.0225	0.01	0.9199
YEAR(OPER)	5	182.9430	36.5886	16.47	0.0001
NEST(PLOT)	42	303.6197	7.2290	3.25	0.0001
ERROR	712	1582.1602	2.2221		

Nested ANOVA for the age at maximum length of ulna

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	0.1582	0.1582	0.001	0.949
PLOT	1	18.6297	18.6297	1.07	0.25
OPER*PLOT	5	177.8620	35.5724	11.99	0.0001
YEAR(OPER)	42	378.7041	9.0168	3.04	0.0001
NEST(PLOT)	1	27.9968	27.9968	9.44	0.0022
ERROR	712	2112.1222	2.9665		

or over time on the same plot. The interaction effects we have detected are in most cases not directly attributable to any one cause. A careful study of the means may allow us to make an interpretation of the interactions.

However, the most we may be able to do is identify a set of factors that could

Table 24. Nested ANOVA for the constant for wing growth in tree swallows.

SOURCE	DF	SS	MS	F	P > F
OPERATION	1	0.00003	0.00003	0.00	0.9547
PLOT	1	0.0007	0.0007	0.18	0.25
YEAR(OPER)	5	0.0374	0.0075	28.26	0.0001
NEST(PLOT)	42	0.0584	0.0014	5.26	0.0001
OPER*PLOT	1	0.0039	0.0039	14.75	0.0001
ERROR	691	0.1827	0.0003		

be causing the interaction. We will probably not be able to single out any factor as the main cause of the interaction.

Statistical Sufficiency -
growth. We have examined the power of each performed test yearly and the difference in means that can be detected with our current data (Zar, 1984, p 260). For the first time we report here AOV tests that incorporate operational period of the antenna, and years along with tests of plots and nests. These new AOV models require a more complex method to ascertain the power of test for each factor and the minimum detectable difference of treatment

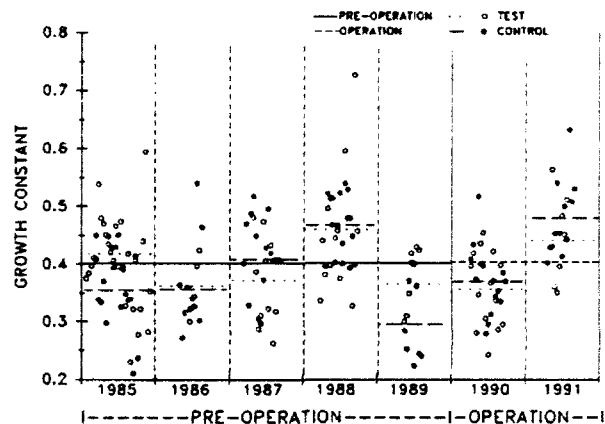


Figure 9. Tarsus growth constants (mm/day) of nestling Tree Swallows observed on test and control sites for 1985 through 1991.

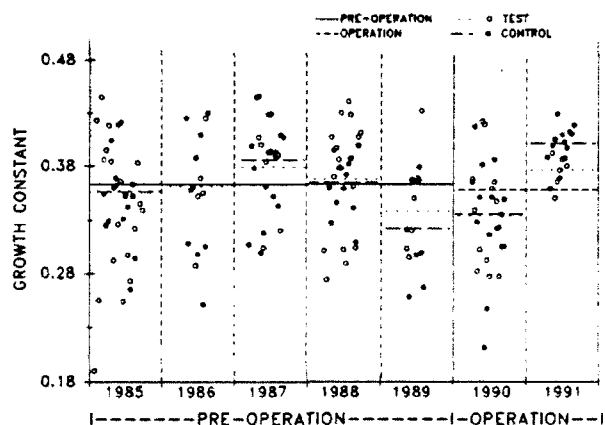


Figure 10. Ulna growth constants (mm/day) of nestling Tree Swallows observed on test and control sites for 1985 through 1991.

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means. We have not as yet satisfied ourselves that we are using the correct power model. Accordingly, we do not present power of test for the full AOV models. We do present power of test results for AOVs done on the curve fit growth variables using our past AOV approach to assure ourselves that no radical changes have occurred. Single AOVs were not calculated for 1991 for linear growth rates, maximum values and maximum ages and we do not present power data for these variables.

Power and minimum detectable values for the curve fit growth variables (Table 25, Table 26) indicate 1991 was similar to other years. Values for wing indicated both power and minimum detectability were better than in previous years. As in previous years, growth constants provide smaller detectable differences than inflection points.

Analysis of Covariance - Growth and Insect Biomass. We attempted the same type of analysis for growth variables as we used for fecundity measures earlier in this report. The index on insect biomass was again the covariate but for this analysis, daily insect biomass was summed for the 15 days preceding hatching (incubation period) to form the covariate INCU and for the

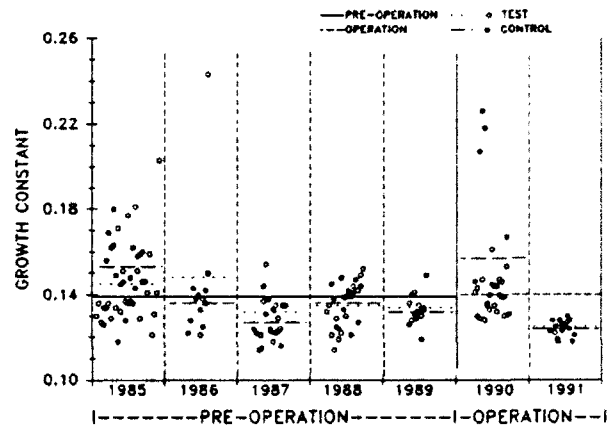


Figure 11. Wing growth constants (mm/day) of nestling Tree Swallows observed on test and control sites for 1985 through 1991.

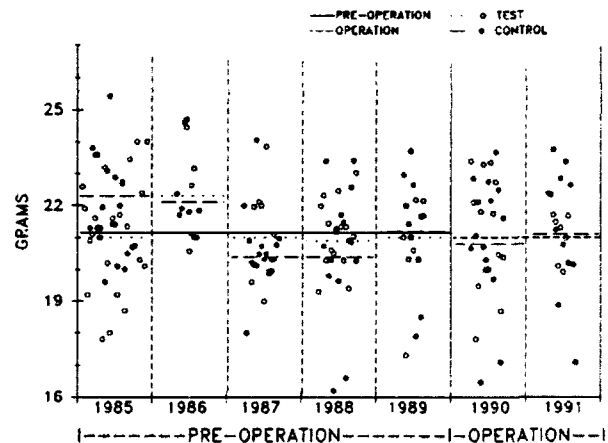


Figure 12. Maximum weight (grams) attained by nestling Tree Swallows observed on test and control sites for 1985 through 1991.

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Table 25. Minimum detectable differences of means for tree swallow growth constants and the minimum percent detectable change in the mean to reach 70% certainty (power) of test. *

Variable	Year	N	Actual Detectable Difference(%)	Actual Power	% Detectable Difference at 70% Power
Weight	1991	8	0.025(4.9)	<.30	0.136(26.7)
	1990	13	0.033(7.3)	<.30	0.125(27.8)
	1989	6	0.058(12.6)	<.30	0.155(33.7)
	1988	13	0.029(6.2)	<.30	0.105(22.3)
	1987	11	0.061(12.7)	<.30	0.118(24.6)
	1986	6	0.087(19.8)	<.30	0.232(52.7)
	1985	21	0.062(13.2)	<.30	0.100(21.3)
Tarsus	1991	8	0.034(7.2)	<.30	0.206(43.8)
	1990	13	0.035(9.5)	<.30	0.128(34.6)
	1989	6	0.125(39.1)	0.40	0.185(57.8)
	1988	13	0.068(14.8)	<.30	0.195(42.4)
	1987	12	0.067(17.2)	<.30	0.150(38.5)
	1986	6	0.072(20.0)	<.30	0.192(53.3)
	1985	21	0.053(13.3)	<.30	0.112(28.0)
Ulna	1991	8	0.045(11.5)	0.62	0.050(12.8)
	1990	13	0.042(12.4)	<.30	0.109(32.1)
	1989	6	0.020(6.1)	<.30	0.126(38.2)
	1988	13	0.038(10.3)	<.30	0.098(26.5)
	1987	11	0.037(9.5)	<.30	0.095(24.4)
	1986	6	0.060(16.7)	<.30	0.160(44.4)
	1985	16	0.052(8.1)	<.30	0.086(23.9)
Wing	1991	8	0.004(3.3)	<.30	0.010(8.3)
	1990	13	0.031(20.7)	.30	0.053(35.3)
	1989	6	0.026(20.0)	<.30	0.068(52.3)
	1988	13	0.008(6.2)	<.30	0.023(17.7)
	1987	11	0.004(3.1)	<.30	0.022(16.9)
	1986	6	0.021(15.0)	<.30	0.085(60.7)
	1985	21	0.006(4.0)	<.30	0.027(18.0)

* The data in this table have been reanalyzed using N = number of nests per treatment and do not agree with figures in earlier annual reports.

15 days following hatching (nestling period) to form the covariate YOUNG. The first step in the analysis was to examine the slopes of the covariates in relation to the main factors in the AOV model: PLOT, OPERATION and YEAR. The regression of the growth variable on the covariate for each main factor must yield lines that are parallel, that is, there must be no interaction between the main factors and the covariate. Significant interaction precludes the use

of the covariate in the analysis.

Unfortunately, we have found significant interactions of the main effects with the covariates INCU and YOUNG for all our growth variables. We therefore cannot perform the analysis that we had hoped would help control for differences in available food among our research plots.

Results of Nestling Transfer

Experiment. Nestling transfers were made in 1990 and 1991 following the methods described earlier. Analysis of Variance was used to examine the effect of TREATMENT with six levels and YEAR with two levels since the experiment was repeated in 1991. The three variables examined were maximum weight, tarsus and ulna because these have the lowest coefficients of

variation of any growth variables and should provide a sensitive test. No effect of the treatment could be detected for any variable (Table 27), nor could we detect differences in various combinations of means in post hoc tests. There was a highly significant year effect due to larger size for all variables in 1990 compared to 1991. This effect was uniform across treatments resulting in no interaction between TREATMENT and YEAR for weight or tarsus. However, a significant interaction was obtained for ulna. The means

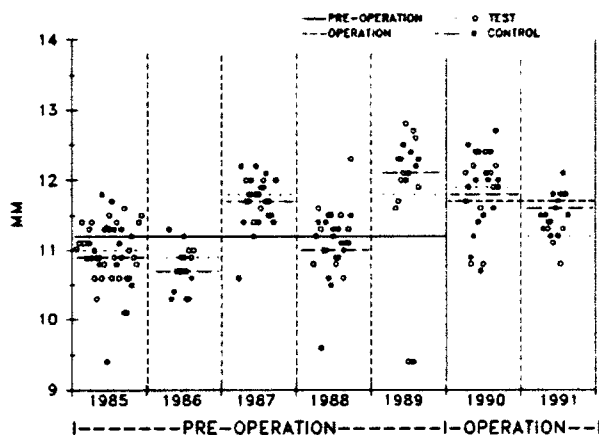


Figure 13. Maximum tarsus length (mm) attained by nestling Tree Swallows observed on test and control sites for 1985 through 1991.

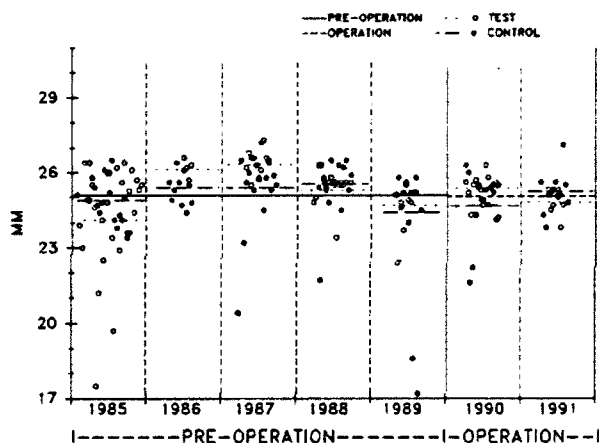


Figure 14. Maximum ulna length (mm) attained by nestling Tree Swallows observed on test and control sites for 1985 through 1991.

Table 26. Minimum detectable differences in mean inflection points and the minimum percent detectable change in the mean to reach 70% certainty (power) of test. ^a

Variable	Year	N ^b	Actual Detectable Difference(%)	Actual Power	% Detectable Difference at 70% Power
Weight	1991	8	1.109(21.7)	0.76	1.02(19.9)
	1990	13	0.656(10.8)	<.30	1.85(30.1)
	1989	6	1.520(24.1)	.40	2.20(34.9)
	1988	13	0.477(8.7)	<.30	1.23(22.4)
	1987	11	0.387(6.8)	<.30	1.95(34.3)
	1986	6	1.090(16.7)	<.30	2.90(44.5)
	1985	21	1.162(19.8)	.59	1.28(21.8)
Tarsus	1991	8	0.460(31.3)	0.30	0.86(58.5)
	1990	13	0.234(10.7)	<.30	0.95(43.6)
	1989	6	1.656(51.8)	.55	1.99(62.2)
	1988	13	0.218(11.6)	<.30	1.03(54.8)
	1987	12	0.619(27.4)	<.30	1.82(80.5)
	1986	6	0.606(31.7)	<.30	1.98(95.2)
	1985	21	0.505(22.4)	.30	0.86(38.2)
Ulna	1991	8	0.884(19.1)	0.65	0.98(21.1)
	1990	13	0.669(12.2)	<.30	1.75(32.0)
	1989	6	1.760(28.9)	.45	2.41(39.6)
	1988	13	0.524(10.6)	<.30	1.45(29.4)
	1987	11	0.447(8.7)	<.30	1.95(37.9)
	1986	6	0.569(9.7)	<.30	2.09(35.5)
	1985	16	0.209(3.6)	<.30	1.54(26.4)

Wing ^a

^a Inflection point not applicable to curves for wing growth.

^b The data in this table have been reanalyzed using N = number of nests per treatment and do not agree with figures in earlier annual reports.

(Figure 16, Figure 17, Figure 15) indicate this interaction was due to the larger means over treatments alternating between 1990 and 1991. Overall, the experiment does not detect any affect on growth due to ELF exposure. It should be noted that the sample N is variable among treatments due to predation (a bear destroyed some nests in 1990) and other accidents. In all treatments with swapped young, the original sample N was 12. In treatments without swapping, the sample N was 34 on control plots and 26 on test plots.

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The high mortality on the test plot was due primarily to predation in both years.

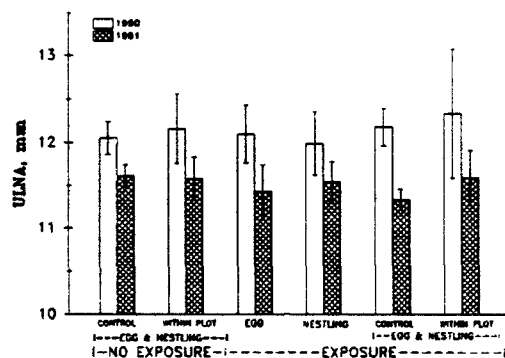


Figure 15 Maximum ulna length (mm) attained by nestling tree swallows during the nestling swap experiments of 1990 and 1991.

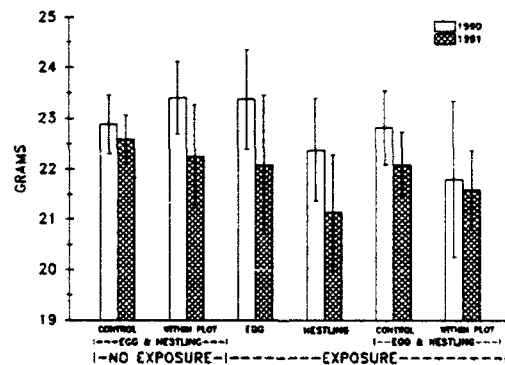


Figure 16. Maximum weight (grams) attained by nestling Tree Swallows observed during the nestling swap experiment conducted in 1990 and 1991.

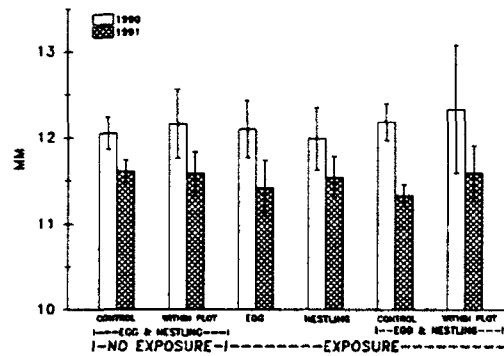


Figure 17. Maximum tarsus length (mm) attained by nestling Tree Swallows observed during the nestling swap experiment conducted in 1990 and 1991.

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Table 27. ANOVA for the nestling exchange experiment done in 1990 and 1991.

MAXIMUM WEIGHT

SOURCE	DF	SS	MS	F	P > F
TREATMENT	5	23.521	4.705	2.051	0.074
YEAR	1	21.962	21.962	9.519	0.002
TREAT*YEAR	5	7.521	1.504	0.656	0.657
ERROR	177	405.962	2.294		

MAXIMUM TARSUS

SOURCE	DF	SS	MS	F	P > F
TREATMENT	5	0.618	0.124	0.612	0.691
YEAR	1	12.502	12.502	61.891	0.0001
TREAT*YEAR	5	1.407	0.281	1.393	0.229
ERROR	177	35.755	0.202		

MAXIMUM ULNA

SOURCE	DF	SS	MS	F	P > F
TREATMENT	5	5.959	1.192	1.487	0.196
YEAR	1	7.099	7.099	8.861	0.003
TREAT*YEAR	5	9.143	1.829	2.282	0.048
ERROR	177	141.817	0.801		

PARENTAL AND NESTLING BEHAVIOR, AND FECUNDITY, GROWTH, AND MATURATION STUDIES - DEERMICE

I. Purpose

The purpose of these studies is to characterize several aspects of the reproductive process in deermice at test and control sites and to test for possible effects of the ELF Communication System on these variables. Specifically, the rates of growth and development of nestlings are compared between test and control sites and for each site from year to year. All of these work elements are described together in this one section because they are all performed on the same families of mice.

II. Methods

These studies were conducted within enclosures because free-ranging mice have been found not to remain resident in nest boxes for long enough periods for us to obtain the data desired. The enclosures are large: 6.1 by 5.8 m. Ten enclosures were constructed within mixed deciduous forests at both the test and control plots. They are open at the top to allow free passage of atmospheric electromagnetic fields and free exposure to weather. Furthermore, they were constructed primarily of acrylic plastic sheeting, which is permeable to atmospheric electric fields according to IITRI engineers. Briefly, the walls of the enclosures consist of acrylic sheeting attached to cedar posts extending about 60 cm above ground and projecting about 15 cm below ground to prevent mice from digging out. A 51-cm-wide sheet of acrylic placed horizontally along the top of each wall prevented animals from climbing over the wall. Tree trunks were sheathed with sheets of high-density polyethylene to prevent mice from climbing in or out of the enclosures via the trees. Each enclosure was provided with a nest box and a feeding and watering station. The nest box can be opened to permit access to the mice.

Small enclosures (termed holding facilities or "hotels") built according to the same design, but measuring just 1.2 by 1.2 m, were also constructed at the same sites. These enclosures were used as holding facilities for mice awaiting study in the large enclosures. The mice to be studied were captured in mixed deciduous forest near the enclosure sites. They were set up as male-female pairs. Later the females were transferred into the large enclosures when visibly pregnant. They gave birth in the enclosures and reared their young to the age of weaning.

Newborn young were toe-clipped for identification when 4 days old. From then until they were 22 days old, their growth was followed by weighing every

other day to an accuracy of 0.1 g using a Pesola scale. Initial litter size and subsequent deaths were recorded. The age of eye-opening and incisor eruption was recorded as an index of developmental rate.

III. Results 1986-1991

The study of growth and development of nestling deer mice was ended with the 1991 season. We have had intractable problems with disease (Tyzzers disease), high female mortality resulting in high death rate among litters where young died due to lack of maternal care, and behavioral problems where females improperly cared for their young or cannibalized them. Efforts to decrease mortality through partial burying of nest boxes to reduce heat stress and keeping the area clean to reduce disease have met with minimal success. We employed extraordinary measures to reduce behavioral problems, all to no avail. Cool weather appeared to substantially increase our chances of obtaining complete data sets on litters dropped in the enclosures, but we had very few seasons that were cool. As a result of these problems, we have a limited data set, and we have relatively poor statistical sufficiency for power of our tests compared to our other tasks.

Growth of Young. A perusal of the growth in body mass of nestlings indicates that growth curves often appear non-linear. Although litter mates consistently exhibit similarly shaped growth curves, there are apparent differences in curves among litters of different females as well as differences between litters of the same female (i.e., some are exponential, some sigmoidal, etc.). While this variability in the shape of growth curves among (but not within litters) is interesting, it precludes the use of any particular non-linear model (e.g., logistic growth model) to estimate and compare growth rates in these mice. Therefore, growth rates have been

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estimated using linear regression analyses for growth of each individual up to the time of weight recession which appears to be correlated with weaning (Figure 18). Analysis of Variance of growth rate with main effects of OPERATION and PLOT, and nested effects due to mothers nested within plot

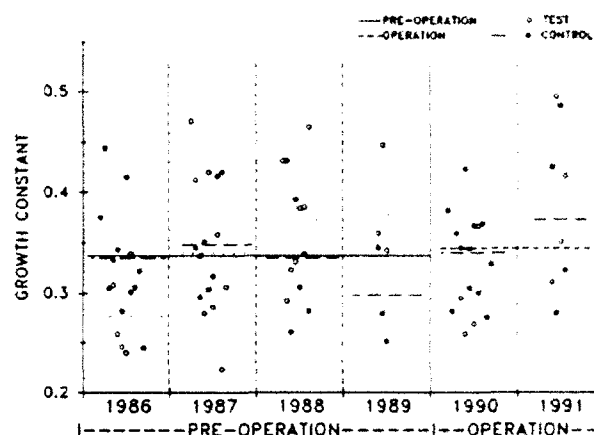


Figure 18. Weight growth constants (grams/day) of young Deermice observed on test and control sites for 1986 through 1991.

Table 28. Analysis of Variance of deermice growth rates on test (Pirilot Road) and control (Michigamme) sites for years 1986 through 1991.

Tested are the effects of PLOT (test vs control), OPERATION (pre- and operational periods) and nested effects MOTHER(PLOT) and YEAR(OPERATION) and interaction terms for 1985 through 1991. See text for method used to determine the proper error mean square to test the main effects in the model.

Source	DF	SS	MS	F-Value	P > F
OPERATION	1	0.0219	0.0219	0.59	0.4838
PLOT	1	0.0005	0.0005	0.008	0.25
YEAR(OPER)	4	0.1472	0.0368	13.83	0.0001
MOM(PLOT)	18	0.4736	0.0263	9.89	0.0001
OPER*PLOT	1	0.0221	0.0221	8.30	0.0042
ERROR	367	0.9761	0.0027		

(MOTHER(PLOT)) and years nested within operation (YEAR(OPERATION)) yielded no significant effects of antenna operation or plot (Table 28). Significant nested effects occurred as did the interaction of operation and plot and plot by year within operation. At this writing, we do not have any hypotheses as to the nature of the mother effect, although it could be related to the number of nestlings in the litter. The year within operation effect seems to be due to the several years of severe drought experienced through 1990. The

interaction terms are more difficult to interpret and require a thorough analysis of the means currently in progress.

Maturation of young. Age at eye opening was over three days earlier at the Pirlot Road test plot in 1991 (Figure 19, Figure 20). Ages at incisor eruption were similar between plots in 1991 as in other years. Age at eye opening and age at incisor eruption was not significantly different among plots or among operation period (Table 29, PLOT,

OPERATION). A highly significant effect due to MOTHER and YEAR was present as were effects due to plot and operation interaction and plot by year within operation interaction for age an eye opening. The latter interaction was not significant for incisor eruption (Table 29). These results parallel those of maturation in the tree swallow. We are currently assessing meaning of the interactions found in the AOV tests.

Statistical sufficiency. The power of the test and the detectable differences were estimated for each

year from 1986 to 1991 (Table 30 - data from 1991 are taken from an AOV done following the same procedures in earlier years, not from the more inclusive

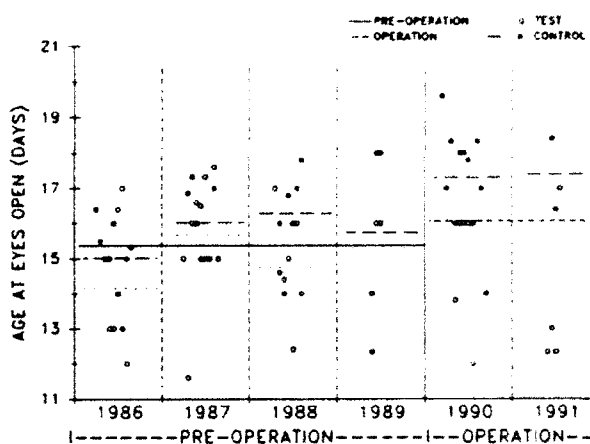


Figure 19. Age at eye opening (days) of young Deermice observed on test and control sites for 1986 through 1991.

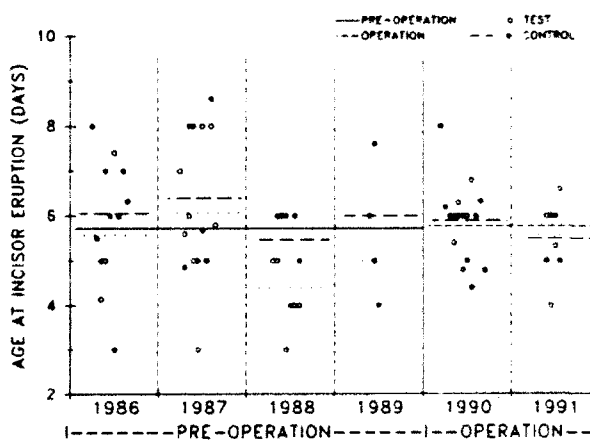


Figure 20. Age at incisor eruption (days) of young Deermice observed on test and control sites for 1986 through 1991.

Table 29. Nested ANOVA of deermice age of eye opening incisor eruption on test (Piriot Road) and control (Michigamme) sites for years 1985 through 1991.

Nested ANOVA of deermice age of eye opening

Source	DF	SS	MS	F-Value	P > F
OPERATION	1	19.6399	19.6399	1.64	0.27
PLOT	1	54.9552	54.9552	2.34	0.10
YEAR(OPER)	4	48.0197	12.0049	6.75	0.0001
MOM(PLOT)	18	332.1698	18.4539	10.38	0.0001
OPER*PLOT	1	99.0215	99.0215	55.69	0.0001
ERROR	345	613.4100	1.7780		

Nested ANOVA of deermice incisor eruption

Source	DF	SS	MS	F-Value	P > F
OPERATION	1	1.7334	1.7334	0.12	0.7447
PLOT	1	0.0251	0.0251	0.004	0.50
YEAR(OPER)	4	56.9469	14.2367	9.74	0.0001
MOM(PLOT)	18	104.1552	5.7864	3.96	0.0001
OPER*PLOT	1	10.4833	10.4833	7.17	0.0078
ERROR	355	519.1465	1.4624		

model presented). The minimum detectable difference ranged from about .11% in 1988 to a high of about 62% in 1989. Minimum detectable differences at 70% power are very large and variable from year to year, much more so than growth for tree swallows. Perhaps this reflects problems in field measurement, but we think it is more a function of the response of the deermice to captivity and handling. They are much more sensitive to handling than the birds.

In general, power of the test for eye opening and incisor eruption were less than or equal to 30% and minimum detectable differences ranged from about 7% to 57% (Table 30). At 70% power, minimum detectable differences vary from about 32% to over 167%. We therefore have relatively poor ability to see small changes in these variables that may result from ELF fields generated by the antenna.

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Table 30. Minimum detectable differences and power for deermice growth constants for years 1986 - 1991

Year	N	Actual Detectable Difference(%)	Actual Power	% Detectable Difference at 70% Power
1991	3	0.154(41.0)	<.30	0.47(125.0)
1990	7	0.051(15.6)	<.30	0.15(45.8)
1989	2	0.212(62.4)	.33	0.34(100.0)
1988	5	0.040(11.4)	<.30	0.24(68.6)
1987	7	0.082(23.6)	<.30	0.22(63.4)
1986	7	0.103(38.1)	.30	0.19(68.5)

Minimum detectable differences and power for deermice maturation events

Variable	Year	N	Actual Detectable Difference(%)	Actual Power	Detectable Difference at 70% Power(%)
Eye opening days (%)	1991	2	8.45(57.0)	<.30	14.3(96.0)
	1990	7	5.4 (32.7)	.74	5.2(31.5)
	1989	2	5.27(34.0)	<.30	16.8(108.4)
	1988	5	3.32(21.4)	.33	5.4(34.5)
	1987	7	1.58(10.0)	<.30	5.0(31.6)
	1986	6	1.00(6.8)	<.30	5.6(38.1)
Incisor eruption days (%)	1991	3	1.55(27.9)	<.30	4.6(82.9)
	1990	7	1.08(18.3)	<.30	2.9(49.2)
	1989	2	2.09(38.0)	<.30	9.2(167.3)
	1988	5	2.42(49.1)	.35	3.5(71.0)
	1987	7	1.54(24.9)	<.30	5.3(85.5)
	1986	6	1.61(27.4)	<.30	4.7(79.7)

Measurement problems. Much of the variation in growth and maturation of young mice can be attributed to the frequency of visits we make to obtain the data (every other day) and also the apparently inherent response to disturbance caused while obtaining measurements. Thus an animal categorized as not having eyes open on a particular day will not be checked again for two days. This produces a built in error of two days. Thus, we do not feel we can obtain fine enough resolution for these variables to meet our statistical criteria without increasing the frequency of visits, yet it is also clear that handling is a major factor affecting growth of the nestlings.

HOMING STUDIES - TREE SWALLOWS

I. Purpose

The purpose of these studies is to measure the homing success of tree swallows at test and control sites and to test for possible effects of the ELF Communication System on such success. Variables measured are the proportions of swallows that successfully return home after displacement from their nest and the time required for each bird to return home. Birds returning to their nest box within 300 minutes from release are considered successful.

II. Methods

Adult birds were captured at the nest box using a passive nest box trapping device (Magnusson 1984). Captures took place between 0800 and 1100 to allow adequate feeding of the young in the nest prior to capture. Following capture, each bird was sexed (using the presence of a cloacal protuberance for males and brood patch for females) and aged using plumage characteristics (Hussell 1983a). Birds were banded using a standard U.S. Fish and Wildlife band and were color marked on the breast using "magic markers" to provide rapid and positive identification while in flight. Birds were placed in wire cages which were covered with black cloths, and then driven to the release sites.

In our first studies of swallow homing in 1984 and 1985, we released birds at all four cardinal compass directions (east, west, north, south) at test and control sites. The results revealed no differences in homing success from one compass direction to another. Furthermore, because tree swallows probably home without regard to habitats they fly over, and they are not likely to be exposed to any different hazards (predators, etc.) in homing from one direction as opposed to another, we feel justified displacing birds in

just one compass direction. This protocol is more efficient in terms of personnel effort than the use of four displacement directions and also permits adequate sample sizes to be obtained.

The release points used for 1986-1990 were located in open areas that are a distance of 30 km from the nest sites and at a compass direction 20 degrees NE of the nest sites (?). This distance was chosen because it is beyond the distance corresponding to a drop of two orders of magnitude of potential electromagnetic fields given off by the Communications System. The direction of the release points in relation to the nest sites was chosen so that birds attempting to return to the nest site in a straight line will cross both east-west legs of the antenna configuration, areas that would supposedly be maximally influenced by ELF electromagnetic fields. Due to reviewer comments on the results from 1986-1990 this protocol was altered for 1991. See the results section below for further details. Upon release, the time, vanishing vector, and weather conditions were noted. Observers located near the nest boxes recorded the time at which the birds returned. Birds at each release site were released singly, with the subsequent bird released when the first had disappeared from sight (approximately three minutes).

III. Results - 1992

Due to prolonged inclement weather from 16-26 June, and overnight freezing weather on 19, 20 and 21 June, we recorded tremendous mortality of young on all plots -- some reaching nearly 100%. Due to this weather-related mortality, very few nests were available from which to home adults. Moreover, when attempts were made to home available adults remaining on the control plot we found that the adult birds were in poor physical condition as evidenced by low body mass. The young in the nest were also of low body mass compared to

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similarly aged young during normal years. In addition, during routine handling and banding of adults to record measures, ages and sexes, we seemingly caused the abandonment of several nests. Handling adults (females in particular) has been shown to cause nest abandonment during egg laying and early incubation, yet rarely during the nestling phase which was when the handling occurred.

Due to lack of available nests, poor weather during homing times, and problems with poor body condition of adults, we did not obtain any tree swallow homing data during 1992. We plan on a final year of data collection during 1993.

Results from the first five years (1986-1990) of the tree swallow homing study have shown that, overall, birds from the test plots are more likely to return than control birds (94.5% return for test birds, 78.7% return for control birds, Table 31). In addition test birds return significantly more rapidly (Figure 21).

In 1990 we attempted to understand these differences without altering our original design, by investigating properties possibly unique to the Panola Plains control

site which could be contributing to the observed differences. We compared the likelihood to return and return speeds for birds displaced from Panola Plains (our normal control birds) to a sample of birds from Tachycineta Meadows control, a site not previously used for homing. Birds from both plots were

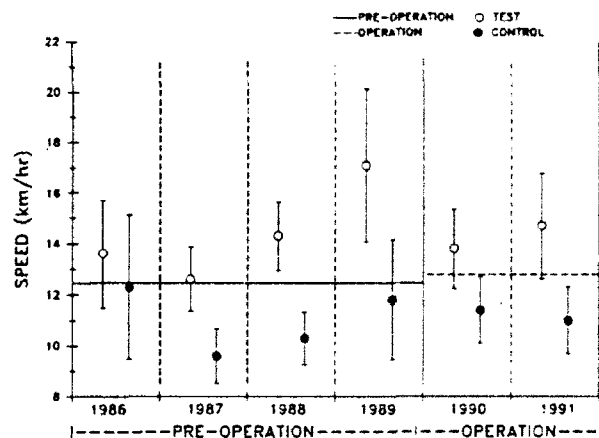


Figure 21. Return speeds (km/hr) of Tree Swallows homed to test and control sites for 1986 through 1991. Yearly plot means (\pm 95% confidence limits) and means for pre-operational and operational antenna status are shown.

Table 31. Results of tree swallow homing study for all years of the study, 1986-1991. Likelihood to return between test and control was tested for each year using a χ^2 test

Year	Plot	Return	Not Return	% Return	χ^2	P
1991	Test	34	1	97.1	13.798	< 0.001
	Control	24	15	61.5		
1990	Test	41	1	97.6	8.004	0.005
	Control	30	9	76.9		
1989	Test	14	0	100.0	0.905	0.341
	Control	15	1	93.8		
1988	Test	37	4	90.2	0.267	0.605
	Control	39	6	86.7		
1987	Test	36	1	97.3	12.258	< 0.001
	Control	25	13	65.8		
1986	Test	26	3	89.7	1.615	0.204
	Control	24	7	77.4		

displaced and released from the normal Panola Plains release site which effectively controlled for any release site characteristics. Likelihood to return was shown to be independent of plot ($G = 2.276$, $P > 0.1$) even though 76.9% returned to Panola Plains while only 53.8% returned to Tachycineta Meadows. The distances travelled by the returning birds were slightly shorter to Tachycineta Meadows (27 km) compared to Panola Plains (30 km), so return speed in km/hour was used for comparison rather than minutes to return. Speeds were shown to be faster for birds returning to Panola Plains (11.4 km/hr) than to Tachycineta Meadows (8.6 km/hr, t-test, $t = 2.037$, $P = 0.049$).

These results suggest differences in plot characteristics between Panola Plains and Tachycineta Meadows rather than a release site effect, yet the number of birds displaced from Tachycineta Meadows represents far too small of a sample to make this analysis conclusive. Even if a larger sample gave the

same conclusion, these results did not help explain the observed differences over five years between test and control plots.

Due to reviewers comments on these 1990 and previous results we changed protocols for the tree swallow homing study during 1991. One of the major criticisms in the past has been the fact that our observed differences may be due to release point differences rather than differences inherent to the test and control plots. In order to test this hypothesis, samples of birds from both test and control would be displaced to their normal release points as well as the other plot's release point. For example, samples of control birds from Panola Plains would be released simultaneously at the normal control release site as well as the test release site. There were several factors which prevented us from using this approach. First, the distances between nest and release point would differ greatly in some cases. The normal displacement distance is 30 km; test birds released at the control point would be displaced 27 km and control birds released at the test point would be displaced 56 km. Secondly, test birds displaced to the control release site would be taken out of the area of electromagnetic influence produced by the antenna, whereas control birds displaced to the test release site would be taken into this area of influence.

As a second choice, reviewers suggested using a common release point for birds displaced from both test and control plots and this was the approach taken. If we followed this protocol and test birds still showed a greater likelihood to return and also returned faster then we could conclude that there was no effect of the release site. When we located on the map a common release point with equal distances to each plot, the site turned out to be within three kilometers of the original control release site. Because of this, it was decided to continue using the same normal control release site as

the common release point even though return distances were 30 km to the test plot and only 27 to the control (see ?). Use of the normal control release point would allow us to continue to compare at least our control data from year to year in an unaltered fashion.

A total of 74 birds were displaced in 1991 (35 test, 39 control) using the common release point. Whereas 97.1% returned on the test plot, only 61.5% returned on the control plot (Table 31). These marked differences in likelihood to return are significant ($\chi^2 = 13.799$, $P < 0.001$) and are similar to results from 1990 and 1987. No significant differences in likelihood to return were found during 1989, 1988, or 1986 (Table 31).

Mean return speed was also shown to be different for the two plots, return speeds for test birds (14.7 km/hr) being significantly faster than control birds (11.0 km/hr, t-test, $t = -3.03$, $P = 0.004$). Faster speeds on the test plots represent a continuation of a trend shown every year of the study to date (Figure 21). These data suggest that release point per se is unlikely a factor in producing the results we have seen thus far, yet the data fail to provide further information explaining observed differences seen consistently throughout the study.

We analyzed data on return speeds from all years of the study (1986-1991) in a nested analysis of variance to assess the potential effects due to antenna OPERATION (preoperational 1986-1989, operational 1990 and 1991), YEAR (nested within operation), PLOT (test vs. control) and OPERATION*PLOT interaction. Due to the nested design, the error term used to compute the OPERATION effect F value was the YEAR (nested within operation) mean square. Results of this analysis (Table 32) show a significant effect due to plot ($F = 38.648$, $P < 0.001$) which is due to return speed being faster on the test plots during every year of the study. Also shown is a significant year effect

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Table 32. Nested ANOVA of tree swallow return speeds, km/hour.

Tested are the effects OPERATION (preoperation 1985-1989, operation 1990-1992), PLOT (test and control), YEAR (nested within operation), and the interaction OPERATION*PLOT. To determine the F value for OPERATION the mean square for YEAR was used in the denominator.

Source	DF	TYPE III SS	MS	F	P
PLOT	1	798.678	798.678	40.32	0.0001
OPERATION	1	0.011	0.011	0.00	0.990
YEAR(OPERATION)	4	236.033	59.008	2.98	0.019
PLOT*OPERATION	1	2.565	2.565	0.13	0.719
ERROR	331	6556.953	19.810		

(F = 2.964 P = 0.020) which is due to return speeds being different to approximately the same magnitude between test and control every year but overall means fluctuating from year to year.

Analyzing the likelihood to home data (Table 31) using the contingency table approach (see tree swallow fecundity section), we first tested for homogeneity of the yearly 2 X 2 tables within operational status. During the four years of preoperational antenna status (1986-1989) the yearly samples were homogeneous ($\chi^2 = 4.578$, df = 3, P > 0.1). The two years of antenna operation (1990 and 1991) were also homogeneous ($\chi^2 = -0.226$, P > 0.5). With data pooled within operational status we tested the hypothesis of mutual independence between likelihood to return, plot (test vs. control) and antenna operation. This hypothesis was rejected ($\chi^2 = 34.47$, df = 4, P < 0.001) leading to the conclusion that some combination of factors influences the rates of return. This led to three tests of partial independence:

1. Likelihood to return is independent of plot and operation.
2. Plot is independent of operation and likelihood to return.

The first two hypotheses were rejected (both $P < 0.001$), but we did not reject the third ($\chi^2 = 4.290$, $df = 3$, $P = 0.232$) and conclude that antenna operation is independent of likelihood to return and plot location. Thus it is desirable to test the hypothesis that likelihood to return is independent of plots location. This was rejected ($\chi^2 = 32.405$, $df = 1$, $P < 0.001$) so we conclude that likelihood to return is highly dependent upon plot location. Overall rates of return have been consistently higher on the test plots (94.9%) compared to the control (75.5%). Throughout the six years of the tree swallow homing study the results have been consistent and straightforward; birds released from the test plots show a higher likelihood to return than control birds, and return speeds are also higher for the test birds. It does not appear that the operational antenna status has had any effect on the results thus far. For a final year of data collection in 1993 we plan on continuing with the common release point approach used in 1991.

HOMING STUDIES - SMALL MAMMALS

I. Purpose

The purpose of these studies is to measure the homing success of small mammals at test and control sites and to test for possible effects of the ELF Communication System on such success. Variables measured are the proportions of individuals that successfully return home after displacement and the time required for each individual to return home. The principal species studied are deermice and chipmunks.

II. Methods

During our initial studies on mammal homing in 1985 (Beaver et al. 1986), we displaced chipmunks and deermice in all four cardinal directions in order to investigate any directional biases in homing ability. No such biases were

found even though animals displaced west and north on the control and test plots had to cross the sham corridor or actual antenna corridor, as well as somewhat different habitat types. However, our sample sizes were small for any particular displacement direction (maximum of 10 animals) and we therefore could not be certain of the robustness of our tests. Thus, in contrast to the work on swallow homing, we decided to reduce the number of displacement directions to two rather than one. Reducing the number of directions from four to two increases efficiency of sampling. By using two directions rather than one, however, we maintained the diversity of habitats and corridor crossings at each site, thus helping to insure that we are further able to examine the effects of habitat conditions as well as potential effects of ELF on homing behavior.

The small mammal homing study was conducted on two trapping grids, one at the Pirlot road test site and the other at the Michigamme control site. Due to the low chipmunk and deermouse populations found in 1985 and 1986, the size of the trapping grid was increased in 1987. Each grid contained 100 stations spaced 15 meters apart rather than ten meters, increasing the area covered to 1.8 ha versus 0.81 ha. One Leathers live-trap was placed at each station, baited with peanut butter and rolled oats. The grids were situated on the east side of both the ELF ROW and the sham ROW. A habitat buffer between each ROW and its respective trapping grid was increased in 1987 to 50 meters, rather than the 10 meters of 1985. This increase helped insure that both the grids and their displacement lines were located in more uniform habitat, a continuous mixed deciduous forest dominated by sugar maple (*Acer saccharum*).

Trapping began on 6 July and ended on 29 July, 1991. Traps were checked twice daily (ca. 0800 and 1900) and re-baited with rolled oats and peanut butter as necessary. Each unique animal was weighed, sexed, and toe-clipped

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upon first capture for individual identification. Reproductive condition, station number, and capture time were also recorded. Individuals were kept for displacement after their third capture; such animals were deemed to be residents of the area where the trapping grid was established which, hopefully, insured their detection by continued recapture on the trapping grid upon returning from displacement. Before being displaced, each animal was kept in a laboratory cage supplied with nesting material, lab chow, and water. Cages were placed in screened-in storage sheds located near each site. Displacements took place during, or just prior to, the next activity period following capture; deermice (nocturnal) were displaced at dusk (ca. 1900) and chipmunks (diurnal) were displaced in the morning (ca. 0800). Each animal was displaced 450 m from the trap it was captured at when kept for displacement. Displacements took place to the south and west of the home grids. The exact point of release was adjusted to reflect the point of capture on the home grid; this way all individuals were displaced exactly the same distance from their capture point. Trapping continued for five days after the last animal was displaced to detect late returns.

The displacements to the south were through continuous forest, whereas those to the west required returning animals to cross the antenna corridor at the test site and the sham corridor at the control site. Use of the two displacement directions thus specifically allowed us to test for directional differences in return rates which might occur due to the fact that animals returning from the west must pass beneath the antenna line, potentially the area of greatest electromagnetic disturbance.

We have experienced varying levels of trap disturbance in past years, primarily at the Michigamme control plot, so we began actively trapping for gray squirrels, skunks, and raccoons in 1992 at the same time we began

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trapping for deermice and chipmunks. We used several sizes of Tomahawk live traps for this purpose. In order to minimize the level of additional predator attraction posed by these traps, we baited these larger traps only with small amounts of peanut butter and rolled oats which is the same bait used in the smaller Leathers traps. Numbers of all three of these species were captured and removed from both test and control plots.

III. Results - 1992

A total of 30 chipmunks (27 test, 3 control) and 23 deermice (12 test, 11 control) were displaced in 1992 (Table 33 and Table 34). No differences in likelihood to return were detected between the two displacement directions for either species, so data from both displacement directions were pooled.

Table 33. Results of chipmunk homing studies at Pirlot Road test plot and Michigamme control plot for all years of the study, 1986-1992

Likelihood to return between test and control was tested for each year using a χ^2 test.

Year	Plot	Return	Not Return	% Return	χ^2	P
1992	Test	19	8	70.4	1.667	0.197
	Control	1	2	33.3		
1991	Test	7	6	53.8	0.151	0.697
	Control	5	3	62.5		
1990	Test	11	12	47.8	2.530	0.112
	Control	15	6	71.4		
1989	Test	15	8	65.2	0.321	0.571
	Control	9	7	56.3		
1988	Test	5	12	29.4	0.200	0.655
	Control	2	3	40.0		
1987	Test	4	8	33.3	0.356	0.551
	Control	2	2	50.0		
1986	Test	13	6	68.4	3.283	0.070
	Control	20	2	90.9		

For deermice, no significant difference in likelihood to return between test and control plots was detected in 1992 ($\chi^2 = 0.157$, $P > 0.9$, Table 34). This is the same result obtained in all years of the study with the exception of 1989 and 1990. There were significant differences in likelihood to return in both 1989 and 1990, although the results for these two years are contradictory to one another. In 1989, a significantly higher percentage of displaced deermice returned to the test plot, whereas in 1990 a significantly higher percentage returned to the control plot.

Considering the years of 1986-1989 as preoperational antenna status and the years 1990-1992 as operational, we tested for heterogeneity of the yearly 2 X 2 tables to determine if these yearly samples could be justifiably pooled within operational status (Zar 1984, pg. 67). See the section on tree swallow

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Table 34. Results of deermouse homing studies at Pirlot Road test plot and Michigamme control plot for all years of the study, 1986-1992

Likelihood to return between test and control was tested for each year using a χ^2 test.

Year	Plot	Return	Not Return	% Return	χ^2	P
1992	Test	9	3	75.0	0.157	0.692
	Control	9	2	81.8		
1991	Test	28	10	73.7	1.462	0.227
	Control	11	8	57.9		
1990	Test	20	23	46.5	10.913	0.001
	Control	29	6	82.9		
1989	Test	13	8	61.9	4.859	0.028
	Control	3	10	23.1		
1988	Test	17	24	41.5	2.181	0.140
	Control	9	5	64.3		
1987	Test	16	7	69.9	0.025	0.874
	Control	6	3	66.7		
1986	Test	5	1	83.3	2.250	0.134
	Control	1	2	33.3		

fecundity for more detailed explanation of this technique.

For deermice during preoperational antenna status it was found that the four years of data (1986-1989) were heterogeneous ($\chi^2 = 8.725$, $df = 3$, $P < 0.05$) and cannot be pooled. During the period of operational antenna status, the same result is obtained; the three years (1990-1992) were found to be heterogeneous ($\chi^2 = 9.09$, $df = 2$, $P < 0.025$) and cannot be pooled.

Since yearly data cannot be pooled to allow testing of preoperational vs. operational conditions another approach is to test the hypothesis of mutual independence (Everitt 1977) within operational status. For deermice during preoperational antenna status we cannot reject the hypothesis of mutual independence ($\chi^2 = 17.22$, $df = 10$, $P = 0.070$) and conclude that the observed likelihood to return is independent of both year and treatment (test or

control plot location). In other words, we found no effect of treatment plot prior to the antenna operation. Considering the antenna operational years of 1990-1992, we reject the hypothesis of mutual independence ($\chi^2 = 17.33$, $df = 7$, $P = 0.015$) and conclude that the observed likelihood to return is due to some combination of year and treatment (test or control plot location). Since we reject the hypothesis of mutual independence we can formulate and test three hypotheses of partial independence (Zar 1984, Everitt 1977) during the years of antenna operation:

1. Return rate is independent of plot and year
2. Plot is independent of year and return rate
3. Year is independent of return rate and plot.

All three of these null hypotheses were rejected (all $P < 0.04$) which leads to the conclusion that during the time of antenna operation (1990-1992), the likelihood of displaced deermice to return is dependent upon year of the sample as well as the treatment plot. This conclusion is reasonable when one looks at the data presented by years in Table 34. The rate of return in 1990 shows a significant difference between test (46.5% return) and control (82.9% return). This result is not repeated in either 1991 or 1992, and moreover, in 1991 the direction of the percentage of returning individuals is opposite that in 1990. Almost equal rates of return were observed in 1992.

Given the results that the present analysis provides and the marked lack of consistency in response between years within antenna operational status, we cannot conclude at this time that the ELF Communications System has an overall effect on deermouse homing, even though there is a significant result in 1990. Further analysis of the present data will continue and we plan on an additional and final year of data collection in 1993.

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For chipmunks, no significant difference in likelihood to return between test and control plots was detected in 1992 ($\chi^2 = 0.417$, $P = 0.519$, using Yates's correction for continuity). This result in 1992 must be tempered by the problem of very low chipmunk numbers on the control plot. These control plot data represent the lowest numbers of individuals displaced in any year of the study (Table 33). All years of the chipmunk study show no differences between test and control plots.

As with the deermice, we tested for heterogeneity of the yearly chipmunk samples within antenna operational status. For chipmunks during the preoperational antenna status, it was found that the four years of data (1986-1989) were homogeneous ($\chi^2 = 0.321$, $df = 3$, $P > 0.9$). The three years (1990-1992) of operational antenna status were also homogeneous ($\chi^2 = 3.924$, $df = 2$, $P > 0.1$), so years were pooled within antenna operation. We then tested the hypothesis of mutual independence on these pooled data and found that the likelihood to return for chipmunks is independent of both treatment plot and antenna operation ($\chi^2 = 5.25$, $df = 4$, $P = 0.264$). We conclude from these results (Table 33) that the likelihood to return for displaced chipmunks is not affected by the ELF Communications System. Further analysis of the present data will continue and we plan on an additional and final year of data collection in 1993.

DEVELOPMENTAL STUDIES

I. Purpose

The purpose of these studies is to characterize aspects of normal embryological development in tree swallows and to investigate potential effects of the ELF communications system on development within the egg. Specifically, early embryological development of tree swallows is being

characterized; developmental abnormalities in field populations of tree swallows are being described and their frequency in test and control plots is being determined, and the size of eggs from test and control plots is being compared.

II. Methods

Tree swallow eggs were collected during late May and early June of 1992. As in previous years, active nests were checked daily and eggs numbered sequentially as they were laid. Five days after the appearance of the last egg, the entire clutch was removed from the nest and coded so that the rest of the analysis could be performed as a blind study. In 1992, 15 clutches were removed from control plots TMC and PPC; 14 clutches were obtained from test plots FST and FNT. Eggs from a sixteenth control nest were not collected because the parents abandoned the nest prior to incubation. A total of 154 eggs was collected of which three were eliminated because they were found to be cracked.

Egg sizes were determined using both weight and volume measurements. Eggs were weighed to two decimal places using a Sybron Digimetric balance. Measurements of egg volumes were obtained by water displacement. Length and breadth were measured using vernier calipers.

All embryos were dissected off the yolk into Howard chick Ringers solution (Johnson and Volpe 1973) and analyzed briefly. The specimens then were fixed in either Bouin's solution or 10% formalin for light microscopy or in 2.5% glutaraldehyde in chick Ringers solution for scanning electron microscopy.

Embryos to be studied with light microscopy were stained as whole mounts using an alcoholic carmine solution (Watterson and Shoenwolf 1984). Following staining, they were dehydrated through a series of graded alcohols and cleared

in methyl salicylate. All embryos were then carefully observed with an Olympus stereomicroscope using transmitted light and a photographic record of any suspected abnormality was obtained. The use of methyl salicylate as a clearing agent allows biological materials to be observed as whole mounts and stored without undue tissue hardening. Subsequently, material can be embedded in paraffin and analyzed by routine histology.

After embryos have been carefully analyzed and a photographed, some are embedded in paraffin, sectioned at 10 microns, and stained with hematoxylin and eosin or toluidine blue for histological examination.

Embryos fixed in glutaraldehyde are dehydrated through a graded series of alcohols, critical point dried and sputter-coated for scanning electron microscopy.

All embryos were staged using the chick embryo series of Hamburger and Hamilton (1951) as a reference. Abnormalities were tabulated and characterized as completely as possible.

This year, in addition to the tree swallow embryos, approximately 100 chick embryos at developmental stages similar to the tree swallow embryos were fixed and treated by the above methods. These provide a solid basis of comparison for the tree swallows which are a previously uncharacterized species.

III. Results

Normal development. As we have reported previously (Beaver, Hill and Hill 1991; Beaver, Hill and Asher 1984), development in tree swallows is similar to that of the chick as described by Hamilton and Hamburger (1951). Although some species differences are apparent and will be briefly mentioned later in this section, comparisons with chick development have been very helpful in determining the "normalcy" of development.

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Tree swallow eggs show an asynchrony of development with the last egg laid routinely lagging behind the rest of the clutch by several stages. We have reported this observation previously (Beaver, Hill and Hill 1991; Beaver, Hill and Asher 1984). In other parts of our study, it is often observed that the last egg hatches approximately a day later than the rest of the eggs. This developmental delay is probably a reflection of the nesting behavior of the parents. Females are frequently observed to spend time on the nest before the clutch is complete; thus, development is probably initiated in "older" eggs before the last egg is deposited. This behavior has been described previously and seems common among small, altricial passerines (Clark and Wilson 1985).

Abnormal development. As previously, embryos were carefully observed for any abnormalities. The status of the following was assessed in each embryo:

Table 35. Frequency of abnormalities found in early tree swallow embryos collected from test and control sites in 1992

Plot	Brain*	Spine	Other	No Development	Total
Test	6 (4)	3 (3)	2 (2)	3 (3)	14 (8)
Control	7 (2)	4 (4)	0	4 (2)	15 (7)
Percent	8.6%	4.6%	1.3%	4.6%	19.2%

* Included in the category of brain abnormalities are 3 embryos from test plots and 6 from control plots which show a flattening of the mesencephalon and a decrease in size and regularity of the cerebral hemispheres. Chi-square analyses are performed both including and excluding these 9 embryos as abnormal. First number is the number of embryos displaying abnormality. Second number (in parentheses) is the number of nests involved.

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developmental stage, brain, eye, ear and branchial arches, heart, spinal cord and somites, limb buds, extra-embryonic membranes and flexion and rotation of the embryo. The results are shown in Table 35. In 1992, 29 embryos or 19.2%, show developmental patterns which fall outside those recognized as normal. Of these, 15 were collected from control plots; 14 were collected from test plots near the communications antenna. (Two eggs from test plot nests and one from a control plot nest were eliminated from the study because they were cracked. The embryos contained were grossly abnormal. We have no certain method of determining cause and effect in these cases, but our experiments in 1990 (Beaver, Hill and Hill 1991) indicated that cracked shells could lead to abnormal development.) No difference in frequency of abnormalities between test and control plots was found (Table 36; Chi-square = 0.0000675, contingency coefficient = 0.000669; $P > 0.9$).

The most frequently occurring abnormalities involved the brain, the spinal column including the nerve cord, notochord and muscle somites, and a failure to develop.

Three embryos, all approximately stage 19, initiated allantois development in wrong direction. These are not included in the "abnormal" group because, as explained previously (Beaver, Hill and Hill, 1991), this problem is hardly ever seen in later embryos and so is probably self-correcting. Consequently it appears more parsimonious to consider it a variation rather than an abnormality. This year, all three embryos came from control plots so there is no evidence that the antenna influences allantois direction.

The most frequently observed abnormality in 1992 involved the formation of the brain. In 3 test plot embryos and 6 control plot embryos the brain was characterized by a pronounced flattening of the mesencephalon and a

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Table 36. Chi-square analysis of developmental abnormalities of embryos including and excluding those with flattened mesencephalon and reduced cerebral hemispheres, 1992

Chi-square analysis of developmental abnormalities of embryos including those with flattened mesencephalon and reduced cerebral hemispheres, 1992.

Status	Normal	Abnormal	Total
Test	59	14	73
Control	63	15	78
Total	122	29	151

Chi-square = 0.0000675

Contingency coefficient = 0.000669

Same test as above but with embryos with flattened mesencephalons and reduced cerebral hemispheres excluded (considered to be a normal variation).

Status	Normal	Abnormal	Total
Test	62	11	73
Control	69	9	78
Total	131	20	151

Chi-square = 0.41

Contingency coefficient = 0.05

irregularity in shape and size of the developing cerebral hemispheres. As with the allantois, this may again be a variation on a theme rather than an abnormality leading to decreased likelihood to survive and reproduce. For this reason, a second calculation of abnormalities eliminating this group was carried out. Elimination of the group reduces the total number of embryos considered abnormal to 20 or 13.3%. Statistical analysis again indicates that there is no difference in frequency of abnormalities between test and control plots (Table 36; Chi-square = 0.41, contingency coefficient = 0.05; $P > 0.1$).

In addition to the 7 embryos which completely failed to develop, two control nests contained embryos which lagged far behind their cohorts. In

both cases, most of the embryos appeared basically normal and these have been included as normal embryos; however, tissue differentiation, particularly of muscle somites, was poor. Whether this is a developmental abnormality or a secondary disintegration resulting from death can not be determined with certainty. We have, in the past, addressed the problem of "parental neglect" (Beaver, Hill and Hill 1991). In 1992, in addition to the two nests just described, a third nest, again from a control plot was observed to be deserted by the parents and those eggs are not included in the sample. In 1991, 3 clutches in which development was severely delayed were collected from test plots; in 1992, 3 delayed and potentially deserted clutches were collected from control plots. We will continue to monitor parental attentiveness but at the moment it does not seem to be linked to the functioning of the communications system.

Our histological investigation of developing tree swallow tissues is ongoing. It is neither practical nor prudent to section all the embryos. We are attempting to section enough normal embryos at each developmental stage to provide a sound reference bank. Some embryos which were detected as abnormal are being sectioned and compared with the normal collection.

We are aware that some abnormalities may only be apparent at the histological level. Accordingly, as few assumptions as possible are made concerning the status of any system. In making our observations, we are paying close attention to the areas where other investigators have reported abnormalities in chick embryos exposed to ELF electromagnetic fields (e.g. nervous system (Ubeda et al. 1992), vestibular ganglia (Leon et al. 1992)).

Reports of adverse effects of ELF electromagnetic fields on development, specifically that of chick eggs exposed experimentally to ELF fields in incubators, continue to accumulate (Juutilanen et al. 1987; Martin 1988, 1992;

Ubeda et al. 1992; Leon et al. 1992; Litovitz et al. 1992). We have suggested that it is possible that chicks and tree swallows have different susceptibilities to ELF fields; however, it seems more likely that the observed differences result from our different experimental designs (Hill et al. 1992). Field strengths and wave forms in the egg/embryo microhabitat probably differ. Because ours is a field study, we are not able to collect these measurements at the egg surface. Moreover, since our experiments are conducted in the field, tree swallow embryos may be partially shielded, at least from electrical fields, by the body of the incubating parent.

With the continued accumulation of data, we will carry out further analyses investigating the possibility that particular types of abnormalities occur with greater frequency in one situation than the other. For example, a vertical analysis before and after the antenna became fully functional of the number of heart malformations or the number of spinal malformations might reveal differences that are masked by an analysis of the total number of abnormalities. To date the numbers of abnormalities in each category is inadequate for such an analysis to be very meaningful.

Size of eggs. Since avian embryos develop in a closed system (the egg), the resources allocated to each offspring by the parent during oogenesis could have a marked influence on the success of the embryo. If females forage less effectively in some situations than in others, eggs may be of lower nutrient value and chick survival may be compromised. To determine whether ELF electromagnetic fields affect the amount of nutrient deposited in eggs, each egg is measured in three ways at the time of collection. First, each egg is weighed. Second, volume is obtained using water displacement. Third, length and breadth of each egg is determined using vernier calipers.

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Weights of eggs from test and control plots have been compared for 5 years before the antenna was fully operational and for 3 years of full operation using nested ANOVAs. Basic statistics and ANOVA results are shown in Figure 22 and Table 37. As noted previously (Beaver, Hill and Hill 1991), nest and year effects are apparent in the data. No difference between test and control plots,

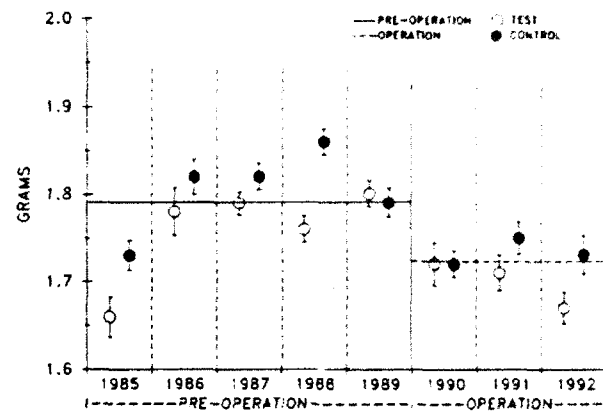


Figure 22. Tree Swallow egg weights (grams) observed on test and control sites for 1985 through 1992.

Table 37. ANOVA of egg weight by PLOT, YEAR, OPERATION and NEST of origin, 1985 to 1992

SOURCE	SUM-OF-SQUARES	DF	MEAN-SQUARE	F-RATIO	P
OPERS	1.133	1	1.133	7.377	0.035
PLOTS	0.257	1	0.257	2.947	0.095
YEARS(OPERS)	0.922	6	0.154	6.592	0.000
NEST(PLOTS)	3.057	35	0.087	3.749	0.000
OPERS*PLOTS	0.004	1	0.004	0.183	0.669
ERROR	26.630	1143	0.023		

before or after the communications system reached full capacity, is seen however. We do see a significant effect due to operation, which we interpret as related to year factors. Since the effect is the same on test and control plots, we do not interpret this as an effect of the antenna.

Because the weight of eggs is affected by both evaporative water loss and metabolic weight loss during development, we have considered it prudent to measure volume to obtain an independent measure of egg size. Prior to the

development of an effective measuring device to determine volume in 1989, the length and breadth of eggs was determined. Since 1990, volume measurements have been obtained in addition to length and breadth (Figure 23). In 1990 and 1991, we used the above measurements to calculate a constant, K , which relates length and breadth to egg volume according to the formula $V_{\text{egg}} = K \cdot B^2 \cdot L$ (Hoyt, 1979) and found no significant difference between K values in test and control plots (Beaver, Hill and Hill 1991). We will now use our calculated K value to estimate volumes of 1992 eggs which will then be compared to measured volumes. If its predictive value is good, K will then be used to calculate egg volumes from the length and breadth data collected prior to 1990. As shown above, year effects are apparent in egg weights, probably resulting in part from different experimental methods, such as slightly different collection times, which could affect weight; therefore, it is of particular interest to see if this difference is reflected in egg volumes.

Comparisons with other species. The development of the domestic chick is the standard against which most other avian species are measured. Early tree swallow development is very comparable to that of the chick although some differences are seen. In 1992, about

100 chick embryos were collected to facilitate the comparison.

Differences between the two species include the constriction of the eye stalk during eye formation, the size of the cerebral hemispheres, the shape of the developing head, the timing of pigmentation of the retina, the relative size of wing buds

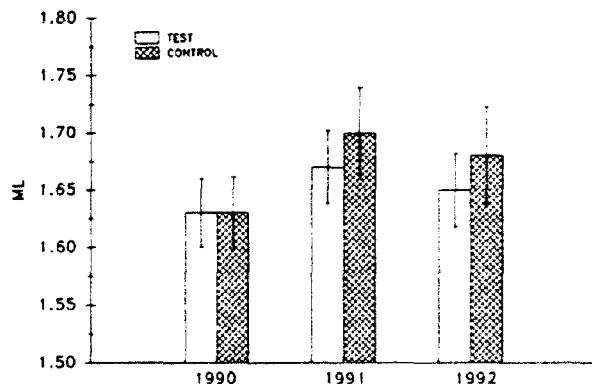


Figure 23. Tree Swallow egg volumes (ml) observed on test and control sites for 1990 through 1992.

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compared to leg buds, and later, the development of feather tracts. A comparison of these differences in development in the two species is underway.

To date we find no difference in the level of abnormalities found in embryos collected from test or control plots. The results of the effects of ELF electromagnetic fields on the embryological development of free-living tree swallows were presented at the First International Congress for Electricity and Magnetism held in June, 1992, at Lake Buena Vista Florida. A paper which will appear in the Proceedings issue is currently in press (Hill, Beaver, Lederle and Herman).

STUDIES OF MAXIMUM AEROBIC METABOLISM

I. Purpose

The purpose of these studies is to measure the peak aerobic metabolism of animals during winter at test and control sites and to test for possible effects of the ELF Communication System on peak metabolism. The principal species studied are chickadees and deermice.

II. Methods

Collection and care of birds. To attract chickadees for study, feeding stations were established in December and kept stocked throughout the winter with sunflower seeds. Chickadees were mist netted as needed from these stations. Upon capture, birds were weighed to the nearest 0.1 g using a Pesola spring scale and marked with a colored plastic leg band for individual identification. When released from captivity, they were banded using a standard U.S. Fish and Wildlife Service band for permanent marking. Birds were housed singly in wire mesh cages (28 x 18 x 31 cm). Shelled sunflower seeds and snow or water were available ad libitum. In addition, each morning and late afternoon, meal worms were provided in excess. The cages were kept

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in a screened outdoor holding facility, which provided natural lighting and temperature conditions.

Collection and care of mammals. Trap shelters were established in late November, prior to any substantial snowfall. The shelters were located along wandering lines situated approximately 75-250 m from the antenna or sham corridor. The habitat was northern hardwoods dominated by maple, basswood, and elm, typical of the area. Each shelter was a plastic waste container placed upside-down on top of the ground layer, with a covered top opening which provided the researcher access to the ground layer once snow was present. Mice entered the shelters through the interface between the ground layer and the wall of the shelter. One Leathers live trap was placed in the bottom of the shelter and baited with rolled oats, peanut butter, and sunflower seeds. Polyester batting was provided in the trap for nesting material. Traps were pre-baited and left open one month prior to actual trapping to insure that small mammals would include the stations in their subnivean runways. Researcher travel on the sites was by snowshoe along a single trail to minimize disturbance of the subnivean air spaces which are critical to small mammal movements.

Trapping was begun at the start of January and continued intermittently, according to need for animals, through March. Work was focused primarily on the deermouse. Upon capture, individuals were toe-clipped for identification, sexed and weighed to the nearest 0.1 g with a Pesola spring scale. Once at the lab, animals were transferred to standard plastic lab cages (29 x 18 x 13 cm) with wire lids and provided with wood shavings, polyester batting, and a diet of sunflower seeds, lab chow, and apple and snow for moisture. Cages were housed in an open outdoor facility which provided natural lighting and temperature conditions.

Laboratory methods. To elicit a peak rate of oxygen consumption, we used a refined version of the helium-oxygen (helox) method first introduced to the study of small-animal physiology by Rosenmann and Morrison (1974). Placing an animal in a helium-oxygen atmosphere at a given ambient temperature greatly increases the individual's rate of heat loss by comparison to the rate in air (mostly nitrogen-oxygen), due to the relatively much higher thermal conductivity of helox. Thus, the animal must produce heat more rapidly in helox than air if it is to maintain a stable body temperature.

Whether the rate of oxygen consumption measured in helox is in fact a true peak metabolic rate depends partly upon the ambient temperature. Identifying the true peak for an individual therefore entails studying the animal at a series of ambient temperatures. Specifically, study at a minimum of three ambient temperatures is required for a definitive determination: there should be a measurement at the temperature that elicits the peak, and also there should be measurements at temperatures higher and lower, demonstrating that the rate of oxygen consumption in helox falls off if the temperature is either raised or lowered from that eliciting the peak. Of course, the temperatures of interest are unknown at the onset of work on an individual. Thus, in principle, many measurements would have to be made on an individual before its peak would be definitively identified. In practice, experience often permits us to know in advance the temperature at which the peak will occur. Therefore, we often need to test an animal at just three temperatures to establish its peak definitively. The spacing we have used between temperatures is 5°C. Thus, if we test an animal in helox at three ambient temperatures that are 5°C apart (e.g. -10, -5, 0°C) and if the highest measured rate of oxygen consumption occurs at the middle temperature, we conclude that we have identified the animal's peak rate definitively.

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Tests were not carried out on the day of capture to reduce any effect of capture stress. To further avoid adverse effects of stress, animals were tested only once on any given day.

Prior to a test animals were weighed to the nearest 0.1 g on an Ohaus triple-beam balance, and their body temperature (T_b) was measured by inserting a copper-constantan thermocouple probe 2-3 cm colonically. Then each animal was placed into a metabolic chamber. Chambers were constructed from new one-half gallon paint cans, with inflow and outflow ports in the lid. The inside surfaces were painted with 3M ECP-2200, for an emissivity of nearly 1.0. A 0.5-inch-mesh hardware cloth floor covered with Dip-It plastic coating was used to elevate the animal above the bottom of the can, thus helping to insure proper airflow around the animal and permitting urine and feces to drop away so as not to wet the animal. The outflow port of each chamber houses a 36-gauge copper-constantan thermocouple to monitor chamber temperature, which is maintained by immersion of the can in a Forma Scientific 2325 water bath using ethanol as antifreeze. All temperature probes are connected to a Leeds and Northrup 250 Series Multipoint recorder which can be read to the nearest 0.1°C.

Measurements were carried out during daylight hours. Food was provided during measurements. Specifically, apple was provided for the mammals, and shelled sunflower seeds and a mealworm were provided for the chickadees. The metabolism chambers for the birds were equipped with a small light that provided dim illumination; without this light, the chickadees (which are diurnal feeders) would not eat. Our decision to provide food during tests is based on extensive preliminary experimentation and is predicated on the following considerations: (1) Animals in nature are able to feed during the day; the birds are diurnal foragers, and the mammals can feed from caches.

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(2) In the mice, the variance in results is lower when food is provided than when it is denied. (3) In the birds, there is evidence that fasting during these types of experiments increases the probability of death.

Oxygen consumption was measured using an open-flow system. Briefly, gas (air or helox) was pumped through the metabolic chamber at a measured flow rate, and the reduction in its oxygen content was measured. From these data, the rate of oxygen use of the animal could be calculated. The oxygen content of gases was measured with an Applied Electrochemistry S3A oxygen analyzer and recorded on a Houston Superscribe potentiometric recorder. Gas flow rates were measured with Brooks 1110 rotameters. The rate of oxygen consumption was calculated according to the formulas in Hill (1972a, method B), taking cognizance of the mathematical relationship between gas composition and the output of the S3A analyzer. We have empirically verified that the S3A analyzer reads oxygen levels in helox with the same accuracy as in air.

Animals were provided with air during an initial adjustment period (0.7-1.5 hr) and then switched to helox. Flow rates were 600 ml/min in air and 900 ml/min in helox. The adjustment period in air was terminated once the metabolic rate remained approximately stable for 15 to 20 minutes. Upon switching to helox, a rapid transition to the new gas was made by purging the metabolic chamber at a rate of 5 liters/min for two minutes. Then the rate of flow was reduced to the 900 ml/min already mentioned. The maximal rate of oxygen consumption under the test conditions was generally achieved within 15-20 minutes after the switch to helox, and animals were rarely exposed to helox for more than 25 minutes. Following the measurement in helox, animals were quickly removed from the metabolic chamber, and a final T_b and weight were recorded. All thermocouples have been calibrated against thermometers whose calibration is traceable to the National Bureau of Standards. Flowmeters have

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been calibrated against a Brooks Volumeter also having a NBS-traceable calibration.

III. Results - 1992

Measures of peak metabolic rate were obtained on 26 deer mice in the winter of 1992. All but two of the measures were obtained within the first week after capture, and the two later measures were obtained within the time period for which we have established that peak metabolic rates are not affected by captivity. Chickadees were not studied in 1992 as part of the planned reduction of effort.

The data were analyzed as specified in the 1988 annual report. It will be recalled that measures of peak metabolic rate are assigned to 10 quality-rating classes. Classes 1, 2, 3, and 4 represent peak determinations of highest quality. Classes 0 and 5-9 represent peak determinations rated as acceptable but nonideal. Statistical comparisons of sets of peak metabolic rates have been carried out using an analysis of covariance design unless otherwise specified. The logarithm of whole-body peak metabolic rate has been used as the dependent variable, and the logarithm of body weight has been used as the covariate. The reasons for the use of analysis of covariance and those for performing the analysis in the logarithmic domain are detailed in the 1988 annual report. Normality of the logarithmically transformed data for peak metabolic rates and body weights was assessed using probit plots, and homogeneity of variances was evaluated with Bartlett's test. Both normality and homogeneity of variances were found to be acceptable in all analyses.

The first step in the analysis of the peak metabolic rates obtained for deer mice in 1992 was to determine if a difference existed between measures that were rated in quality classes 1-4 (primary quality) and those that were rated in the other quality classes (secondary quality). This was done by

Table 38. Summary of peak metabolic rates measured on deer mice soon after capture in 1992. All measured peaks, regardless of their quality rating, are included

Plot	Number of Measures	Peak Metabolic Rate [ml O ₂ /(g X hr)]		Mean Body Weight (g)
		Mean	S.D.	
MGE (Control)	11	18.3	1.6	19.1
PRT (Test)	15	17.5	2.1	20.1

pooling all data from both test and control plots into an analysis of covariance with a single factor: primary versus secondary quality rating. As in all past years, the difference between the quality rating categories for deer mice proved nonsignificant ($P = 0.27$). Thus, for analysis of plot effects in 1992, all peaks were pooled regardless of their quality rating. A single-factor analysis of covariance was performed on these pooled peaks, the factor being plot (test versus control). The effect of the covariate (body weight) was not significant ($P = 0.15$). Moreover, there was no significant difference between test and control plots ($P = 0.82$). Summary statistics are given in Table 38. We conclude that for deer mice in 1992 peak metabolic rates measured soon after capture did not differ between the test and control plots.

Summary of data for preoperational and fully operational years. As documented in the annual report for 1990, we have data for both deer mice and chickadees during two winters in which the ELF Communications System was not operational at all: 1986 and 1987. For chickadees we acquired data for two winters in which it was operational at full power and in modulated mode for most of the time: 1990 and 1991. The chickadee data for 1986-87 (preoperational years) and 1990-91 (fully operational years) were summarized

Table 39. Summary of peak metabolic rates measured on deer mice soon after capture in 1986-87 and 1990-92. All measured peaks, regardless of their quality rating, are included

Years and Plots	Number of Measures	Peak Metabolic Rate [ml O ₂ /(g X hr)]	
		Mean	S.D.
1986-87			
MGE (Control)	18	20.2	1.6
PRT (Test)	17	19.2	1.6
1990-92			
MGE	34	18.6	1.7
PRT	38	18.2	2.1

and compared in the annual report for 1991 and are not repeated here. For deer mice, the data gathered in 1992 complete the data set and provide us with results for three winters in which the Communications System was operational at full power and in modulated mode. Table 39 summarizes the data on deer mice for 1986-87 (preoperational years) and 1990-92 (fully operational years). All peak metabolic rates measured for deer mice are included, regardless of quality class, because in all the years concerned there was no significant difference between data placed in different quality classes.

The summary data in Table 39 have been analyzed by two-way analysis of covariance. The dependent variable was whole-body peak metabolic rate. The factors were time (1986-87 versus 1990-92) and plot (test versus control). As described above, the analysis was carried out in the logarithmic domain. In this domain, the pooled data are beautifully normal and variances are robustly homogeneous. The data set was unbalanced. Thus, each of the two possible hierarchical (sequential) analyses of covariance was carried out (time first, plot second; plot first, time second). The two hierarchical analyses agreed in statistical outcome.

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The two time periods differed ($P = 0.002$ in both hierarchical analyses). Rates tended to be lower in 1990-92 than in 1986-87. The two plots did not differ ($P = 0.07-0.09$), and there was no significant interaction between time and plot ($P = 0.78$). Thus, there is no evidence that operation of the Communications System has altered the peak metabolic rates of deer mice. The reason for the overall decrease in rates on both plots between 1986-87 and 1990-92 is unknown.

Overall, we have found no effect of the operation of the Communications System on the peak metabolic rates of either chickadees (1991 annual report) or deer mice (present report).

OVERALL RESEARCH FINDINGS

In conclusion, we have made our first statistical appraisals accounting for pre- and operational status of the antenna, test and control plots, years of data collected in the periods of antenna operation and nests on the plots. We have also completed analyses of covariance using insect biomass as a factor in measures of fecundity and growth, and of initial body mass in the metabolism of deermice and chickadees. We also report on a two-year experiment where we exchanged nestlings of tree swallows among test and control plots. All of these studies were directed to an assessment of the effect of the ELF antenna system on the various variables we measure on our study animals. Our findings to date show, for the most part, that operation of the antenna system has not measurably changed the fecundity, mortality, growth and maturation or development of our study animals. We have also not found an antenna effect on small mammal or tree swallow homing, or peak metabolism of deermice or chickadees.

We plan careful examination of future data to further assess these findings. In other cases, we have found differences with no discernible relation to the antenna. We will continue to evaluate these findings as well.

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